CLINICAL TRIAL PROTOCOL

Protocol Number	IEDAT-04-2022
Trial Title	A Multi-center, Randomized, Double-blind, Placebo-controlled Trial to Evaluate the **N**eurological Effects of **E**ryDex on subjects with **A**taxia-**T**elangiectasia
Short Trial Title - Acronym	**NEAT trial**
Phase	Phase 3
EUCT Number	[redacted]
IND Number	[redacted]
Date of Protocol	Version 5.0 EU: 22-AUG-2025
Author	[redacted]
Sponsor	Quince Therapeutics S.p.A. Via Meucci, 3 20091 Bresso (MI), Italy [redacted]

Good Clinical Practices Statement

This trial will be performed in compliance with Good Clinical Practices, the Declaration of Helsinki (with amendments) and local legal and regulatory requirements.

Quince Therapeutics S.p.A., Via Meucci 3, 20091 Bresso (MI), Italy

1 SYNOPSIS

Title of Study	A Multi-center, Randomized, Double-blind, Placebo-controlled Trial to Evaluate the ***N****eurological* Effects of ***E****ryDex* on subjects with ***A****taxia-****T****elangiectasia* (***NEAT*** trial)
Protocol #	IEDAT-04-2022
Phase of Development	3
Planned Center(s)	Approximately 20 centers
Planned Trial Period	First Participant In: Q2 2024 Last Participant In: Q3 2025 Last Participant Last Visit: Q4 2025
Study Objectives	**Primary Efficacy Objective:** To evaluate the effect of EryDex (encapsulated dexamethasone sodium phosphate [eDSP]) on central nervous system symptoms, as measured by the change of the rescored modified International Cooperative Ataxia Rating Scale (RmICARS) from Baseline to Visit 9 compared to placebo in ataxia-telangiectasia (A-T) (6- to 9-year-old participants primary analysis population). [*The RmICARS is a re-scored version of the International Cooperative Ataxia Rating Scale (ICARS) and consists of 9 items across 3 domains (kinetic domain and speech limited to one item each), totaling a maximum score of 29 points.]* **Key Secondary Efficacy Objective:** To evaluate the effect of eDSP compared to placebo, in A-T (6- to 9-year-old participants primary analysis population), based on: • Change of Clinical Global Impression of Severity (CGI-S) from baseline to Visit 9. **Other Secondary Efficacy Objective:** • Clinical Global Impression of Change (CGI-C) at Visit 9. **Exploratory Objectives:** To evaluate the effect of eDSP on quality of life (QoL), compared to placebo, in A-T (6- to 9-year-old participants primary analysis population), based on: • Health-related quality of life (QoL) using the change from baseline to Visit 9 of the domains in the EuroQoL 5D-Y (EQ-5D-Y) Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older • Health-related QoL using the change from baseline to Visit 9 of the EQ-5D-Y total score and visual analogue scale (VAS). **Participants Aged 10 Years and Older:** These primary, secondary, and exploratory efficacy objectives will also be evaluated for the participants aged 10 years and older. These analyses will be performed as data summaries in this older age cohort. **Safety Objective:** To evaluate the safety and tolerability of eDSP compared to placebo in participants with A-T, based on the occurrence of Treatment-Emergent Adverse Events (TEAEs), including serious adverse events (SAEs) and discontinuations due to AEs, and changes in vital signs, laboratory parameters, electrocardiograms (ECGs), and physical/neurological examination findings. Safety analysis will be performed on the total population, on the 6- to 9-year-old participants, and on participants ≥10 years old.

Study Design	This is an international, multi-center, randomized, prospective, double-blind, placebo-controlled, Phase 3 trial, designed to assess the effect of eDSP (dexamethasone sodium phosphate [DSP] in autologous erythrocytes), administered by intravenous (IV) infusion once every 28 days (window +2 days, -7 days), on neurological symptoms of participants with A-T. The primary analysis population includes children 6 to 9 years old. Participants 10 years old and above will also be enrolled. The EryDex System (EDS) is a drug-device combination product that is used to load DSP into autologous erythrocytes creating eDSP (previously referred to as EryDex), which is infused into the participant. [redacted] Upon completion of all screening assessments for eligibility, participants meeting all selection criteria at baseline will be randomized in a 1:1 fashion to eDSP or placebo. A total of 86 participants 6 to 9 years old, approximately 43 per group, will be randomized. To ensure the enrolment of the primary analysis population, the total number of participants 10 years of age and above is limited to 1 per site without prior approval from Sponsor (approximately 20 participants 10 years of age and above, 10 per treatment group, may be enrolled). The randomization will be stratified by age (6 to 9 years old and ≥10 years old), sex (male, female), and by geographic region. Participants will be considered to have completed the trial when Visit 9 has been performed. An open-label extension (OLE) trial will be offered to all participants who complete the full treatment period, complete the final trial assessments, do not present safety contraindication to continuation of treatment, and who provide informed consent. The ICARS will be administered by a blinded, qualified rater per site (neurologist initially identified by the Principal Investigators (PIs) based on experience and then qualified and trained specifically for the administration of the ICARS in the IEDAT-04-2022 trial). The ICARS rater will not be involved in the rating or administration of the CGI-S, CGI-C, QoL, Columbia-Suicide Severity Rating Scale (C-SSRS), or in the safety assessments and cannot access the safety data. The ICARS rater will not be allowed to administer the trial treatment. The ICARS ratings must be completed without consulting scores from the previous visit. The rater for CGI must be a physician familiar with the participant but does not need to be a neurologist. The rater for CGI (not allowed to administer the trial treatment and blinded to safety assessments and safety data as well) will not have access to the ICARS ratings. The CGI rater will be initially identified by the PIs based on experience and then qualified and trained specifically for the administration of the CGI in the IEDAT-04-2022 trial. The CGI rater must review and consult the following tools to perform an assessment of various aspects of the participant's presentation and provide CGI scores: baseline CGI-S assessment video and notes (when scoring CGI-C). At each applicable visit, the ICARS should be the first scale administered, followed by the CGI--S/CGI-C, and then by the EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older (please refer to Table 1 for the list of tests required at each visit). Neurological assessments must be performed before any phlebotomy or IV insertion so that upper extremity neurological exam is not impeded by an IV line, except at screening and Visit 9.
Planned Number	A minimum of 86 participants 6 to 9 years old, and approximately 20 participants aged 10 years and older, meeting all selection criteria, will be enrolled and randomized to 1 of the 2 treatment

of Participants	groups (approximately 43 participants per group aged 6 to 9 years, and 10 participants per group in aged 10 years and older).
Inclusion and Exclusion Criteria	**Inclusion Criteria** 1. Participant meets clinical criteria for diagnosis of A-T. The neurological signs of A-T (incoordination of the head and eyes in lateral gaze deflection, gait ataxia associated with an inappropriately narrow base) must be documented. Such signs of A-T illustrate the body systems in which changes shall be confirmed, but the listed signs are examples, and other changes in those systems may be observed and documented to confirm the diagnosis of A-T. 2. Participant is in autonomous gait or is helped by periodic use of a support (i.e., ICARS score for *Item 1 – Walking Capacities* between 0 and 4 included). 3. Participant is at least 6 years of age (Dose 1 must be on or after date of 6th birthday), of either sex. 4. Genetic confirmation of A-T. 5. Body weight ≥15 kg. 6. The participant and parent/caregiver (if below the age of consent), or a legal representative, has provided written informed consent to participate. If consent is provided solely by the caregiver in accordance with local regulations, the participant must provide assent to participate in the trial, to the extent possible. **Exclusion Criteria** General 1. A disability that may prevent the participant from completing all trial requirements. 2. Current participation in another clinical trial. Participation in observational, non-interventional studies is allowed with approval by the Medical Monitor as long as trial investigational endpoint raters can remain blinded to the assessments from other studies, and as long as the other trial participation does not interfere with participation in this trial. Medical History and Current Status 3. Immune impairment that includes cluster differential 4 positive (CD4+) lymphocytes count <400/mm^3 (for participants less than 7 years old) or <150/mm^3 (for participants ≥7 years old). In the presence of oral infections, like oral candidiasis, documented at the screening or recurrent as per medical history documentation, the limit increases to <200/mm^3 (for participants ≥7 years old). 4. History of severe impairment of the immunological system such that steroid treatment would be contraindicated. 5. Loss/removal of 250 mL or more of blood within the past 4 weeks prior to screening. 6. Current neoplastic disease or previous neoplastic disease not in remission for at least 2 years. 7. Severe or unstable pulmonary disease that impacts participant participation in the trial, in the opinion of the Investigators. 8. Uncontrolled diabetes. Participants with diabetes that has been stabilized (i.e., no hypoglycemic or hyperglycemic episodes in the past 3 months) will be eligible. 9. Any other severe, unstable, or serious disease or condition that in the Investigator's opinion would put the participant at risk for imminent life-threatening morbidity, need for hospitalization, or mortality. 10. Any clinically significant abnormality on standard laboratory examinations (hematology, biochemistry, urinalysis) at screening that remains abnormal on repeat testing, if considered as a possible sign of a clinical condition putting the participant at risk if enrolled. Eligibility of participants with abnormal laboratory test values will be determined by the Investigator in consultation with the Medical Monitor. 11. Participant with an early morning plasma cortisol level below 3-5 µg/dL (depending on assay), or participant exhibits signs or symptoms of adrenal insufficiency (Appendix 7), with

	an early morning plasma cortisol level below 10 µg/dL, and fails the adrenocorticotropic hormone (ACTH) stimulation test at screening. 12. Confirmed hemoglobinopathies, e.g., hemoglobin C disease, sickle cell anemia, hereditary spherocytosis, or thalassemia). 13. Current chronic or acute significant renal and/or hepatic impairment that in the Investigator's opinion will impact participant participation in the trial. 14. Participants with suicidal ideation. 15. Females who are pregnant or breast feeding. Females of childbearing potential using an adequate birth control method, as determined by their healthcare provider, will be eligible. For further details on adequate contraceptive measures, please refer to Section 14.7. Prior/Concomitant Medication 16. Any previous oral or parenteral steroid use within 6 weeks before Baseline. Treatment with inhaled or intranasal steroids for asthma or allergies, as well as use of topical steroids will be permitted. 17. Chronic condition or prior allergic reaction representing a contraindication to the use of dexamethasone or other steroid drugs. 18. Has participated in any other trial with an investigational drug and received a dose within 30 days or less than 5 half-lives (whichever is greater) prior to the Screening Visit. 19. Has participated in a previous trial with EryDex treatment. 20. Requires any concomitant medication prohibited by the protocol, please refer to Section 11.5.
Schedule of Visits and Assessments	
Screening Period	Prior to the initiation of any screening procedure, every participant and parent/caregiver (if below the age of consent), or a legal representative must provide written consent and sign the informed consent form; in accordance with local regulations, children must provide assent to participate in the trial, to the extent possible. During the screening period, any previous treatments with other corticosteroid compounds will be withdrawn (washout from previous treatment); such washout from previous treatment may (depending on the dose of the previous corticosteroid) result in a prolongation of the standard 30-day screening period to 45 days from the completion of the tapering off, which, in this specific case, will not constitute a deviation from the protocol. Following informed consent, Investigators should consult with the Medical Monitor about such specific cases. The following screening evaluations will be conducted: • Medical history and demographics • Physical/neurological examination • Vital signs (including height and weight) • C-SSRS • Review of eligibility criteria • ICARS (Note: the full ICARS exam will be done at each assessment. The RmICARS will be calculated as part of the data analysis) • 12-lead standard ECG • Routine laboratory tests: hematology, biochemistry (including serum creatinine), urinalysis • Female participants will have a serum pregnancy test • Special laboratory tests: CD4+ lymphocytes count, alpha-fetoprotein, and C-reactive protein (CRP) • Plasma cortisol – sample to be collected before 8:00 AM and prior to randomization at baseline. If the basal cortisol level is within the reference normal range, the participant can be enrolled in the trial. If the 8:00 AM cortisol level is below 3-5 µg/dL (depending on assay) regardless of symptoms, or if the participant exhibits signs or symptoms of adrenal insufficiency (Appendix 7) and has a cortisol <10 µg/dL, the participant will receive a high-

	dose ACTH stimulation test as soon as possible. If the ACTH stimulation test is normal, the participant can be enrolled after the effects of the ACTH have dissipated (e.g., 30 days after ACTH administered, resulting in a prolongation of the screening period). Potential participants failing the ACTH stimulation test will be excluded from the trial and referred to an endocrinologist (a pediatric endocrinologist, depending on the participant's age) with a recommendation to prescribe stress dose steroids • Assessment of prior and concomitant medications • Assessment of AEs occurring after giving informed consent, during the screening period, and before the first dose of trial treatment If any abnormal laboratory test results, vital sign measurements, or ECG findings of clinical significance are noted at the screening visit, these must be repeated during the 30-day screening period and the results made available prior to making the final decision on a participant's eligibility for the trial at Baseline. Adverse events (AEs), reported by the participant or observed by the Investigators, and the use of concomitant medication will be recorded from the time of signing of informed consent through the end of the trial. Participants who initially fail eligibility criteria for the trial may be re-screened in cases where the PI, in consultation with the Medical Monitor, believes this is appropriate based upon an understanding of the health condition of the participant.
Visit 1 Baseline and Dose 1	The following assessments will be performed, and trial eligibility will be confirmed before administration of trial treatment: • Neurological examination • Review of all eligibility criteria • ICARS • CGI-S (videotaped) • EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older • C-SSRS • 12-lead standard ECG (only repeat if abnormal findings at the Screening Visit) • Routine laboratory tests on the diverted blood sample (only repeated due to abnormal parameters at the Screening Visit) • Special laboratory tests on the diverted blood sample: CD4+ lymphocytes count and CRP • Assessment of physical development, sexual maturation, and the effect of trial treatment with dexamethasone on these aspects (Tanner scale; Marshall and Tanner 1969, 1970[1]). Tanner staging of the breasts in premenarchal females and of the scrotum in males who have not completed puberty, will be performed • Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to infusion • Bone mineral density, where country regulations allow • Vital signs, within 15 minutes before the IV infusion of the trial treatment • Assessment of concomitant medications • Assessment of AEs that occurred before the first dose of trial treatment • Blood sample for biomarker development on the diverted blood sample (if specific consent is provided)

[1] Marshall WA, Tanner JM (June 1969). Variations in pattern of pubertal changes in girls. Arch. Dis. Child. 44 (235): 291–303. Marshall WA, Tanner JM (February 1970). Variations in the pattern of pubertal changes in boys. Arch. Dis. Child. 45 (239): 13–23.

	Following randomization using Interactive Web Response System, trial treatment will be prepared and administered immediately after completion of the EryDex process, as follows: • [redacted] blood collected after blood diversion (see Section 11.3) for aerobic culture • [redacted] blood collected after blood diversion and sampling for culture, for use in the EryDex process • Addition of randomized trial medication (DSP or placebo) to the EDS • Upon completion of the EryDex process, fill the satellite sample bag with [redacted] • Trial treatment administration by IV infusion, TO BE STARTED within 30 minutes after the EDS process has completed • Once the final bag is empty (EryDex has been infused), flush the infusion lines with normal saline only as per clinical practice (e.g., by infusion pump or by gravity) • The satellite sample bag will be detached and used for sterility culture tests: a sample of EryDex (approximately [redacted]) will be collected to perform a sterility test using a culture-based method as presented in Appendix 5. [redacted] sample of EryDex will be stored under refrigeration as a "Retention Sample"." • In case of positive/contaminated post-release sterility results, an action plan has been established as presented in Appendix 6. The remaining sample in the satellite sample bag or, if this is not available, a sample collected from another EryDex sampling point, will be used for determination of DSP content and complete blood count (CBC), as presented in Appendix 3 After completion of the IV infusion of the trial treatment, the following assessments will be performed on Day 0: • Vital signs, within 15 minutes after the infusion • Assessment of concomitant medications • Assessment of AEs Physical examination must be conducted on the day of trial treatment administration.
Visit 2	This visit should be performed remotely, by phone, unless there are specific safety/medical concerns that would prompt an onsite visit. This remote safety follow-up should be performed within 24 hours (allowed window: + 2 days) after the first dose of trial treatment. At Visit 2 the following assessments will be performed: • Assessment of concomitant medications • Assessment of AEs
Visit 3	This visit should be performed remotely, by phone, unless there are specific safety/medical concerns that would prompt an onsite visit. At Visit 3 (allowed window: ±3 days) the following assessments will be performed: • Assessment of concomitant medications • Assessment of AEs
Visits 4, 5, 7, and 8	At Visits 4, 5, 7, and 8 the following assessments will be performed: • Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to each infusion • C-SSRS • Vital signs, within 15 minutes before the IV infusion of the trial treatment • Assessment of concomitant medications • Assessment of AEs

	The trial treatment will be prepared and administered immediately after the completion of the EryDex process, as follows: • [redacted] blood collected after blood diversion (see Section 11.3) for aerobic culture • [redacted] blood collected after blood diversion and sampling for culture, for use in the EryDex process • Addition of randomized trial treatment (DSP or placebo) to the EDS • Upon completion of the EryDex process, fill the satellite sample bag with approximately [redacted] • Trial treatment administration by IV infusion, TO BE STARTED within 30 minutes after the EDS process has been completed • Once the final bag is empty (EryDex has been infused), flush the infusion lines with normal saline only as per clinical practice (e.g., by infusion pump or by gravity) • The satellite sample bag will be detached and used for sterility culture tests: a sample of EryDex [redacted] sterile sample of EryDex will be stored under refrigeration as a "Retention Sample" • In case of positive/contaminated post-release sterility results, an action plan has been established as presented in Appendix 6. The remaining sample in the satellite sample bag or, if this is not available, a sample collected from another EryDex sampling point, will be used for determination of DSP content and CBC, as presented in Appendix 3 After completion of the IV infusion of the trial treatment, the following assessments will be performed: • Vital signs, within 15 minutes after the infusion • Assessment of concomitant medications • Assessment of AEs Physical examination must be conducted on the day of trial treatment administration.
Visit 6	At Visit 6, the following assessments will be performed before trial treatment administration, unless specified otherwise: • Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to infusion • Neurological examination • ICARS • CGI-S, CGI-C • EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older • C-SSRS • Routine laboratory tests (on the diverted blood sample) • Special laboratory tests (on the diverted blood sample): CD4+ lymphocytes count, alpha-fetoprotein, and CRP • Blood sample for biomarker development on the diverted blood sample (if specific consent is provided) • Vital signs, within 15 minutes before the IV infusion of the trial treatment • Assessment of concomitant medications • Assessment of AEs

	The trial treatment will be prepared and administered, immediately after the completion of the EryDex process, as follows: • [redacted] blood collected after blood diversion (see Section 11.3) for aerobic culture • [redacted] blood collected after blood diversion and sampling for culture, for use in the EryDex process • Addition of randomized trial treatment (DSP or placebo) to the EDS • Upon completion of the EryDex process, fill the satellite sample bag with approximately [redacted] • Trial treatment administration by IV infusion, TO BE STARTED within 30 minutes after the EDS process has been completed • Once the final bag is empty (EryDex has been infused), flush the infusion lines with normal saline only as per clinical practice (e.g., by infusion pump or by gravity) • The satellite sample bag will be detached and used for sterility culture tests: a sample of EryDex [redacted] sterile sample of EryDex will be stored under refrigeration as a "Retention Sample" • In case of positive/contaminated post-release sterility results, an action plan has been established as presented in Appendix 6. The remaining sample in the satellite sample bag or, if this is not available, a sample collected from another EryDex process sampling point, will be used for determination of DSP content and CBC, as presented in Appendix 3 After completion of IV infusion of trial treatment, the following assessments will be performed: • Vital signs, within 15 minutes after the infusion • Assessment of concomitant medications • Assessment of AEs Physical examination must be conducted on the day of trial treatment administration.
Visit 9/Early Withdrawal/ Trial Completion	At Visit 9, the following assessments will be performed: • Physical/neurological examination • Vital signs • ICARS • CGI-S, CGI-C • EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older • C-SSRS • 12-lead standard ECG • Assessment of physical development, sexual maturation, and the effect of the administered treatment on these aspects. Tanner staging of the breasts in pre-menarchal females and of the scrotum in males who have not completed puberty will be performed • Females noted to be at Tanner Stage 2 or greater at this visit will have a serum pregnancy test • Routine laboratory tests • Special laboratory tests: CD4+ lymphocytes count, alpha-fetoprotein, and CRP

	• Plasma cortisol – sample to be collected before 8:00 AM. If the 8:00 AM cortisol level is below 3-5 µg/dL (depending on assay) regardless of symptoms, or the participant exhibits signs or symptoms of adrenal insufficiency (Appendix 7) and has a cortisol <10 µg/dL, the participant will receive a high-dose ACTH stimulation test as soon as possible. Participants failing the ACTH stimulation test will be referred to an endocrinologist (a pediatric endocrinologist, depending on the participant's age) with a recommendation to prescribe stress dose steroids • Bone mineral density, where country regulations allow • Assessment of concomitant medications • Assessment of AEs • Blood sample for biomarker development (if specific consent is provided) Participants who complete the full treatment period, complete the trial assessments, and provide informed consent will be eligible to receive treatment with EryDex in an OLE trial. Participants who discontinue prematurely will still be asked to perform this visit.
Unscheduled/ As-needed Assessments	Participants in whom there is a clinical suspicion of hemolysis during the trial may have the following testing performed depending on the clinical circumstances and as determined by the Investigators: • Hemolysis Panel: free plasma hemoglobin, haptoglobin, bilirubin (total and conjugated), CBC, lactate dehydrogenase (LDH) and urinalysis • Red blood cell (RBC) antibodies (immunoglobulin [Ig]G, IgM, Qualitative Direct Coombs test) • Free plasma hemoglobin (1 hour after infusion) In addition, Investigators can decide to perform any additional safety assessments deemed necessary to assess participants' safety throughout the trial.
Investi-gational Medicinal Product(s): dose, mode of admini-stration, and dosing schedule	The trial treatment consists of DSP administered via ex vivo encapsulation into autologous erythrocytes, or an infusion of erythrocytes prepared using a placebo solution instead of a DSP solution, that are infused immediately after encapsulation into the same participant with A-T. The EDS is used to load DSP or placebo solution into autologous erythrocytes, which are infused into the participant. Dexamethasone sodium phosphate administered IV is progressively dephosphorylated by the enzyme phosphatase, normally present within RBCs, to dexamethasone, with kinetics dependent on the quantity of substrate present. Thus, intra-erythrocyte phosphatases play a critical role in delivering the final, free dexamethasone. The progressive dephosphorylation is very fast in the first 24 to 48 hours and much slower over the following 30 days. Autologous whole blood [redacted] to be used for the EDS process will be collected by peripheral venipuncture into heparinized collection tubes. Eligible participants will be randomized to receive trial treatment with encapsulated DSP (Group 1) or Placebo (Group 2). Group 1 will receive an IV infusion of [redacted] of encapsulated DSP. The infusion will be a total volume of approximately [redacted] (final volume of the bag minus the volume transferred into the satellite sample bag) and will be given over approximately 40 minutes. Group 2 will receive an IV infusion of encapsulated Placebo in a total volume of approximately [redacted] (final volume of the bag minus the volume transferred into the satellite sample bag) given over approximately 40 minutes. Trial treatment will be administered by IV infusion every 28 days calculated from the date of the first dose. There is an allowed window of -7 to + 2 days for each treatment visit. The window between an infusion and the subsequent one should be kept as regular as possible throughout the

	trial, avoiding fluctuations in administration windows. In any case, infusions should not be given fewer than 14 days apart. Any instances where it is not possible to administer the infusion within the designated window should be documented as a protocol deviation and the Investigators should immediately contact the Medical Monitor to discuss if the participant may continue in the trial, and if so, to agree on the path forward that will best ensure the participant's safety and meet the protocol goals. [redacted] will be provided in 10-mL amber-colored [redacted] for participants assigned to Group 2. The ex vivo encapsulation of the trial treatment (DSP or placebo solution) into autologous erythrocytes will be performed using the EDS by means of the Red Cell Loader, a single-use, disposable kit (EryKit_01), the Syringe Kit, and process solutions, which are all Conformité Européenne (European Conformity; CE)-registered Medical Devices, according to manufacturer instructions. [redacted].
Reference Therapy	For participants randomized to the Placebo group, [redacted] not containing DSP will be used in the EDS process.
Planned Treatment Duration per Participant	The trial will consist of a 30-day screening period followed by an approximately 6-month treatment period. Participants who complete the trial's full treatment period, and complete the trial assessments may elect to receive treatment eDSP in an OLE trial.
Analysis Populations	The following analysis sets will be used: • *Intent-to-treat (ITT) Population*: the ITT Population is defined as all participants who were randomized. • *ITT (6-9) Population*: the ITT (6-9) Population is based on the ITT Population and comprises randomized participants aged 6 to 9 years. This population will be the one on which main efficacy analyses are conducted. • *ITT (10+) Population*: the ITT (10+) Population comprises randomized participants aged 10 years and older. • *Per-Protocol Population (PP)*: a subset of the ITT (6-9) Population comprising all participants aged 6 to 9 years who qualified for the ITT (6-9) Population, met trial treatment adherence requirements, completed Visit 9, and did not have disqualifying protocol deviations as specified in the Statistical Analysis Plan. Classification of participants in the PP Population will be done in a blinded manner before database lock. • *Safety Population (SP)*: any participant who received at least 1 dose of the randomized treatment.
Statistical Methods (including sample size calculation)	The sample size for this trial is based on a comparison between active and control in the change from baseline to Visit 9 for the RmICARS. The null hypothesis will be no difference in the change from baseline RmICARS between active and control, with the alternative hypothesis of a difference of -2.4 between active and control. A total sample size of 86 participants 6 to 9 years old (43 per treatment group) will provide approximately 90% power with a two-sided test at 0.05 type I error, assuming an SD of 3.0 for the treatment group and 3.7 for the control group. The number of aged 10 years and older is not based on the ability to detect a specific treatment effect.

	Efficacy Analysis The primary efficacy endpoint, the change from baseline to Visit 9 in RmICARS in participants aged 6 to 9 years old, will be analyzed using a mixed-model repeated-measures analysis, as it allows for data missing at random (MAR) to be imputed from this model. Data missing for reasons other than treatment discontinuation due to AEs or deaths will be imputed in this way. The analysis will be conducted by imputing datapoints after death to be the worst value for the endpoint at that visit across the trial, and by imputing datapoints after treatment discontinuation due to AEs using a Jump to Reference approach, which assumes the RmICARS score behavior for participants withdrawing due to AEs is the same as in the control group. The analysis model will include the change from baseline to Visits 6 and 9 as the repeated measure, with treatment as the fixed effects and terms for region, sex, and visit, and baseline RmICARS, sex, and age as continuous covariates, and the treatment-by-visit interaction. The treatment effect, together with the associated 2-sided 95% CI and p-value, will be calculated. Unstructured covariance matrices will be used. Model effect estimation will be based on restricted maximum likelihood. Sensitivity analysis using a retrieved dropout imputation method for discontinuation due to AEs will be performed to address possible issues related to missing data. **Secondary Endpoints** A hierarchical testing procedure will be used to control for multiplicity to maintain overall alpha at 0.05. The following secondary endpoints will be formally tested in order only if the primary analysis of RmICARS showed $p < 0.05$ **Key Secondary Endpoint:** The change in CGI-S will be considered the key secondary trial outcome. The change in CGI-S from baseline to Visit 9 will be classified as improved versus no change/worsened. The proportion improved will be analyzed with logistic regression, with covariates for age, baseline RmICARS, sex, and region. **Other Secondary Endpoint:** The difference in the outcome on the CGI-C ordinal scale at Visit 9 between treatment groups will be analyzed with a Proportional Odds logistic regression model. The resulting proportional odds ratio represents the likelihood of participants in the eDSP group being in a better level of the CGI-C compared to the participants in the placebo group. The proportional odds model will include covariates for baseline RmICARS, age, sex, and region. Summary statistics will be used to show the proportion of participants in each level, as well as the individual odds ratios at each level. Should a level contain fewer than 5 participants across treatments, or only 1 participant in either treatment arm, the level will be collapsed to the next higher (worse) level. Within the range of responses, the proportion of participants who report any improvement (levels 1-3, included) versus those who report no change or worsening (levels 4-7, included) will be of primary interest. If the proportional odds assumption is violated, a logistic regression based on the proportion improved (Levels 1-3) versus no change and worsened (Levels 4-7) will be used. The logistic regression model will include covariates for age, baseline RmICARS, sex, and region. **Exploratory Endpoints** EQ-5D-Y Interviewer Administered Proxy version 1 will be used for participants 6 to 9 years of age and self-reported for participants 10 years of age and older, with each of the 5 domains evaluated, and a total score calculated. VAS will be reported separately on a scale from 0 to 100. The change from baseline to Visit 9 in total score and VAS will be analyzed with Analysis of Covariance, with baseline score, region, sex, baseline RmICARS, and age as covariates. As a supplemental analysis, the proportion of participants who do not worsen in total score and VAS will be compared between the 2 treatment arms with logistic regression. The logistic regression model will include covariates for baseline score, region, sex, baseline RmICARS, and age.

	Safety Analysis The safety and tolerability of eDSP compared to placebo in participants with A-T, based on the occurrence of TEAEs, including SAEs and discontinuations due to AEs, and changes in vital signs, laboratory parameters, ECGs, and physical/neurological examination findings, will be assessed by the incidence of TEAEs, which will include SAEs, and events by severity and relationship to treatment. Changes in clinical laboratory results and vital signs will be presented, along with summaries of clinically significant changes. All AEs experienced after receiving treatment will be considered as TEAEs.. Safety analysis will be performed on the total population, on the participants aged 6 to 9 years old and on participants aged ≥10 years old. **Data Safety Monitoring Board** An independent Data Safety Monitoring Board (iDSMB) will review safety and tolerability data from the trial. The iDSMB will receive unblinded safety data for review at specified intervals and may recommend to modify or to stop the trial if significant safety concerns are detected. The iDSMB will review all the safety data on an ongoing basis, with special emphasis on the incidence and severity of steroid-related events, new infections, SAEs, and deaths, in addition to the standard safety parameters.

Table 1: Schedule of Visits and Assessments

Visit	**Screen**	**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	**V7**	**V8**	**V9**
		Treatment Period								**Trial Visit 9 (b, c)**
Dose		**Dose 1 Baseline(a)**	**(d)**	**(d)**	**Dose 2**	**Dose 3**	**Dose 4**	**Dose 5**	**Dose 6**	**Trial Completion /Early Withdrawal**
Visit Window			**24 hours after V1**	**Day 14 (±3 days)**	Each treatment must be scheduled every 28 days (window -7 days, +2 days,), calculated from the previous visit					28 days (window -7 days, +2 days), calculated from the previous visit
Informed Consent	X									
Medical History/Demographics	X									
Inclusion/Exclusion Criteria	X	X								
EryDex Infusion		1			2	3	4	5	6	
Neurological Examination	X	X					X			X
Physical Examination	X	X			X	X	X	X	X	X
Tanner Scale (p)		X								X
Vital Signs, Height, Weight	X	X (l)			X(l)	X(l)	X(l)	X(l)	X(l)	X
Early morning plasma cortisol test (h)	X	As needed								X
ECG	X	X(e)								X
Routine Laboratory Tests (f)	X	X(e)					X			X
BMD, where countries allow		X								X
ICARS	X	X					X			X
C-SSRS (j)	X	X			X	X	X	X	X	X
Special Laboratory Tests (g)	X	X					X			X
Pregnancy testing (n)	X	X			X	X	X	X	X	X
Quality of Life EQ-5D-Y (q)		X					X			X
CGI-C							X			X
CGI-S (videotaped at V1)		X					X			X
Hemolysis Panel (k)	As needed									
RBC antibodies (k)	As needed									
Free plasma hemoglobin (k)	As needed									
Blood sample for biomarker development (o)		X				X				X
Pre-dose aerobic blood culture (m)		X			X	X	X	X	X	
Culture-based sterility test on EryDex (m)		X			X	X	X	X	X	
EryDex sample for CBC, DSP content (i)		X			X	X	X	X	X	

Visit	**Screen**	**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	**V7**	**V8**	**V9**
		Treatment Period								**Trial Visit 9 (b, c)**
Dose		**Dose 1 Baseline(a)**	**(d)**	**(d)**	**Dose 2**	**Dose 3**	**Dose 4**	**Dose 5**	**Dose 6**	**Trial Completion /Early Withdrawal**
Visit Window			**24 hours after V1**	**Day 14 (±3 days)**	Each treatment must be scheduled every 28 days (window -7 days, +2 days,), calculated from the previous visit					28 days (window -7 days, +2 days), calculated from the previous visit
Prior/Concomitant Medications	Throughout the duration of the trial									
Adverse Events	Throughout the duration of the trial									

Abbreviations: ACTH = adrenocorticotropic hormone; AE = adverse event; BMD = bone mineral density; CBC = complete blood count; CGI-C = Clinical Global Impression of Change; CGI-S = Clinical Global Impression of Severity; C-SSRS = Columbia-Suicide Severity Rating Scale; D = Day; DSP = dexamethasone sodium phosphate; ECG = electrocardiogram; EQ-5D-Y = EuroQol 5D-Y; ICARS = International Cooperative Ataxia Rating Scale; SAE = serious adverse event; V = visit.

Notes:

Each treatment must be scheduled every 28 days (window +2 days, -7 days), calculated from the previous infusion. The window between an infusion and the subsequent one should be kept as regular as possible throughout the trial, avoiding fluctuations in administration windows. In any case, infusions should not be given fewer than 14 days apart.

Any instances where it is not possible to administer the infusion within the designated window should be documented as a protocol deviation and the Investigators should immediately contact the Medical Monitor to discuss if the participant may continue in the trial, and if so, to agree on the path forward that will best ensure the participant's safety and meet the protocol goals.

Unless specifically described, the trial visits must be performed on site. Under exceptional/specific circumstances (such as public health reasons, participant's illness), remote visits can be allowed on a case-by-case basis, if onsite visits are not allowed. In such instances, the Investigators should immediately contact the Medical Monitor to discuss the visit's conduct. Any missed procedure that cannot be done remotely will be identified as a protocol deviation.

At each applicable Visit, the ICARS should be the first scale administered, followed by the CGI-S/CGI-C, and then by the EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and the self-reported version for participants ≥10 years of age. Neurological assessments must be performed before any phlebotomy or intravenous insertion so that the upper extremity neurological exam is not impeded by an intravenous line, except at screening and Visit 9.

Footnotes:

a) Baseline visit is designed as a single-day visit but, in case of logistical issues for a particular participant, the visit can be performed over a 2-day period. In this case, the day of the first dose will be considered as Baseline, and subsequent treatments will be calculated from the previous dose.
b) Efficacy evaluations (Visit 9) will be used as the Endpoint assessments for all efficacy measures.
c) Participants who discontinue prematurely will still be asked to perform this visit. Participants who complete the trial's full treatment period and complete the trial assessments may elect to receive treatment with eDSP in an OLE trial..
d) Visits 2 and 3 should be performed remotely, by phone, unless there are specific safety/medical concerns that would prompt an onsite visit.
e) These evaluations will be repeated only if abnormalities requiring follow-up were noted at the Screening evaluation; results from the repeat assessments must be available at baseline to confirm eligibility before the participant can be randomized to treatment. If the Screening ECG is found to have abnormalities requiring follow-up, it will be repeated (in triplicate) at Baseline and evaluated to determine eligibility prior to randomization.
f) Routine laboratory assessments to include complete hematology, biochemistry (including serum creatinine), and urinalysis.

g) Special laboratory tests include CD4+ lymphocytes count, α–fetoprotein (not repeated at baseline), and C-reactive protein.
h) A sample will be collected before 8:00 AM for measurement of plasma cortisol during the screening period, prior to randomization at baseline. If the basal cortisol level is within the reference normal range, the participant can be enrolled in the trial. If the 8:00 AM cortisol level is below 3-5 µg/dL (depending on assay) regardless of symptoms, or the participant exhibits signs or symptoms of adrenal insufficiency (see Appendix 7) and has a cortisol <10 µg/dL, the participant will receive a high-dose ACTH stimulation test as soon as possible. If the ACTH stimulation test is normal, the participant can be enrolled after the effects of the ACTH have dissipated (e.g., 30 days after ACTH administered, resulting in a prolongation of the screening period). If the participant fails the ACTH stimulation test, they will be excluded from the trial treatment and from continuing further participation in the trial (this is not applicable at V9 testing) and referred to an endocrinologist (a pediatric endocrinologist, depending on participant's age) with a recommendation to prescribe stress dose steroids. In addition to the testing during the screening period (prior to randomization), a blood sample to be collected before 8:00 AM for measurement of plasma cortisol in the following instances: (1) when participants are symptomatic, and (2) when participants are at most risk for adrenal insufficiency and adrenal crisis, such as following discontinuation or interruptions of dexamethasone dosing (i.e., loading failures and following discontinuation of the trial treatment).
i) Upon completion of the EryDex System process, the remaining sample in the satellite sample bag or, if this is not available, a sample collected from another EryDex sampling point, will be used for determination of DSP content and CBC.
j) C-SSRS: to be administered at all the visits. "Baseline-Screening" version at Screening and "Since Last Visit" version at all subsequent visits.
k) Participants in whom there is a clinical suspicion of hemolysis during the trial may have the following testing performed depending on the clinical circumstances and as determined by the Investigators: hemolysis panel [free plasma hemoglobin, haptoglobin, bilirubin (total and conjugated), complete blood count, lactate dehydrogenase, and urinalysis], red blood cell antibodies (immunoglobulin G, immunoglobulin M), Qualitative Direct Coombs test, and Free Plasma Hemoglobin (1 hour after infusion).
l) Vital signs to be measured within 15 minutes before and within 15 minutes after infusion at Visits 1, 4, 5, 6, 7, and 8. Weight and height to be measured just once per treatment visit and before the infusion.
m) [redacted]
n) Female participants will have a serum pregnancy test obtained at Screening. Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated before each infusion at Visits 1, 4, 5, 6, 7, 8 and a serum pregnancy test at Visit 9/Early Discontinuation, if noted to be at Tanner Stage 2 or greater at this Visit.
o) For participants who have provided specific consent.
p) Should the Principal Investigator/delegated Investigator note evidence, or should the parent report any evidence of female trial participants approaching puberty (e.g., breast development, pubic hair, or onset of menses) after the Baseline and during the trial, an ad hoc full Tanner staging will be performed to ensure female participants will start pre-treatment pregnancy testing, as needed.
q) EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older.

2 SIGNATURE PAGE

Trial Title	A Multi-center, Randomized, Double-blind, Placebo-controlled Trial to Evaluate the **N**eurological Effects of **E**ryDex on subjects with **A**taxia-Telangiectasia
Trial Code	IEDAT-04-2022
Protocol Version/Date	Version 5.0 EU – 22-Aug-2025
Number of Centers	Approximately 20
Protocol Authors	[redacted]
Sponsor Representative	[redacted] [redacted] 01-Sep-2025 | 16:15:46 PDT _____ _____ Date [redacted] the design of the trial

Clinical Research Organization

The Sponsor has transferred all responsibilities for the conduct of the trial to the Contract Research Organization or other designated vendors for specific services.

3 TABLE OF CONTENTS

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4 ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation	Explanation
ACTH	adrenocorticotropic hormone
AE	adverse event
AESI	adverse events of special interest
A-T	ataxia-telangiectasia
ATM	ataxia-telangiectasia mutated
ATP	adenosine triphosphate
BARS	Brief Ataxia Rating Scale
BMD	bone mineral density
BMI	body mass index
BP	blood pressure
bpm	beats per minute
CBC	complete blood count
CD4+	cluster differential 4 positive
CE	Conformité Européene
CFR	Code of Federal Regulations
CGI-C	Clinical Global Impression of Change
CGI-S	Clinical Global Impression of Severity
CNS	Central Nervous System
COPD	chronic obstructive pulmonary disease
^{51}Cr	chromium-51
CRF	Case Report Form
CRO	Contract Research Organization
CRP	C-reactive protein
C-SSRS	Columbia-Suicide Severity Rating Scale
CYP3A4	cytochrome P450 3A4
D	Day
DSP	dexamethasone sodium phosphate
ECG	electrocardiogram
EDC	electronic data capture
EDS	EryDex System
eDSP	encapsulated Dexamethasone Sodium Phosphate
EQ-5D	EuroQol 5-dimension
EQ-5D-5L	EuroQol 5 dimension 5 level
EU	European Union
FAS	Full Analysis Set (Population)
FDA	Food and Drug Administration
GCP	Good Clinical Practices
HDL	high-density lipoprotein
IB	investigator's brochure
IBD	inflammatory bowel disease
ICARS	International Cooperative Ataxia Rating Scale
ICF	Informed Consent Form

Abbreviation	Explanation
ICH	International Conference on Harmonisation
iDSMB	Independent Data Safety Monitoring Board
IEC	Independent Ethics Committee
Ig	immunoglobulin
IM	intramuscular
IMP	investigational medicinal product
IND	Investigational New Drug (application)
IRB	Institutional Review Board
ITT	Intent-to-Treat (Population)
IV	intravenous
IWRS	Interactive Web Response System
IU	international unit
LDH	lactate dehydrogenase
MAR	missing at random
mICARS	Modified version of the International Cooperative Ataxia Rating Scale excluding oculomotor items (17-19) and items 8-12
OLE	open-label extension
PI	Principal Investigator
PK	pharmacokinetic(s)
PP	Per Protocol (Population)
QoL	quality of life
RBCs	red blood cells
RCL	red cell loader
RmICARS	Rescored Modified version of the International Cooperative Ataxia Rating Scale
SAE	serious adverse event
SAP	Statistical Analysis Plan
SARA	Scale for Assessment of Rating of Ataxia
SD	standard deviation
SP	Safety Population
T50	time to disappearance of 50% of the labelled red blood cells from the circulation
TEAEs	treatment-emergent adverse events
US	United States
VABS	Vineland Adaptive Behavior Scale
VAS	visual analogue scale
vs	versus
WBC	white blood cells

5 TITLE OF TRIAL

A Multi-center, Randomized, Double-blind, Placebo-controlled Trial to Evaluate the Neurological Effects of EryDex on subjects with Ataxia-Telangiectasia.

6 PROTOCOL NUMBER

This trial is being conducted under Protocol No. IEDAT-04-2022.

7 BACKGROUND AND TRIAL RATIONALE

7.1 Background Information

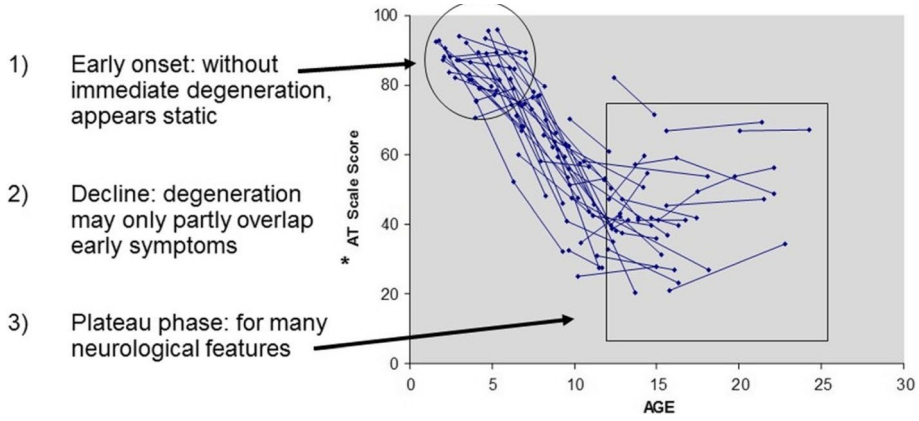
Results derived from the completed Phase 3 IEDAT-02-205 (ATTeST) trial were used to inform the design of a new, adequate, and well-controlled trial to evaluate the efficacy and safety of EryDex for the treatment of ataxia-telangiectasia (A-T).

IEDAT-04-2022 is an international, multi-center, randomized, prospective, double-blind, placebo-controlled, Phase 3 trial, designed to assess the effect of EryDex (DSP in autologous erythrocytes), administered by intravenous (IV) infusion once every 28 days (window +2 days, -7 days), on neurological symptoms of participants with A-T. Approximately 86 participants between 6 to 9 years old will be enrolled and analyzed as the primary analysis population; approximately 20 participants ≥10 years old will also be enrolled.

Based on the natural history data and results from ATTeST, the selection of the 6- to 9-year-old population for this Phase 3 trial may allow for clear separation from placebo and demonstration of a treatment effect over a 6-month treatment period.

Natural history data (Crawford et al., 2006; Rothblum-Oviatt et al., 2016; Jackson et al., 2016) show that rapid deterioration of motor functions is observed before age 10. By the age of 12, the vast majority of A-T participants become wheelchair-dependent and the neurological signs of disease progression slow significantly primarily affecting upper limb and respiratory functions. As shown in Figure 1, participants <6 years of age show limited clinical signs of neurological deterioration, while between 6 to 10 years of age, there is a rapid clinical deterioration, which subsequently plateaus.

Figure 1: Disease Progression (A-T-Nest) Versus Age (Rothblum-Oviatt et al., 2016)



Abbreviations: A-T = ataxia telangiectasia; AT = ataxia telangiectasia

Similar results were obtained from longitudinal data collected from 35 A-T participants (Jackson et al., 2016).

These natural history data are supported by a cross-sectional analysis of the baseline RmICARS scores; an identical pattern can be observed using Modified International Cooperative Ataxia Rating Scale (mICARS) or full International Cooperative Ataxia Rating Scale (ICARS) scores from a large cohort of A-T participants (n = 261) from the ATTeST trial, the Phase 2 IEDAT- ERY01-2010 trial and a non-interventional trial (Nissenkorn et al., 2015). As shown in Figure 2, [redacted]

In the ATTeST trial, there was a much larger EryDex treatment effect in the 6- to 9-year-old subpopulation. These findings are aligned with the natural history data that show a slow rate of decline for ataxia symptoms up to age 4 to 5 years, increasing from age 6 to 9 years and then decreasing significantly from age 10 years onwards.

The safety profile of the EryDex treatment did not raise clinical concerns about the overall safety in any of the studies conducted to date in A-T participants, nor concern regarding the known long-term side effects of chronic steroid treatment in A-T population, regardless of the age.

Given these results, trial IEDAT-04-2022 is being conducted to develop EryDex as a clinically meaningful treatment for these patients, who suffer from a devastating progressive disorder for which there are currently no viable treatment options.

7.1.1 *Overview of the EryDex System*

The EryDex System (EDS) incorporates (encapsulates) dexamethasone sodium phosphate (DSP) into autologous red blood cells (RBCs), allowing the slow release of low doses of dexamethasone (active drug) in circulation (plasma) (Mambrini et al, 2017).

[redacted]

Dexamethasone is well suited for this type of application. Dexamethasone sodium phosphate is biologically inactive and once embedded within the RBCs does not diffuse out (Magnani et al, 1998; Castro et al, 2007; D'Ascenzo et al, 1997; Crinelli et al, 2000). [redacted]

[redacted] The enhanced penetration of the central nervous system (CNS) by dexamethasone compared with prednisolone (Mitchell et al, 2005), together with the low plasma exposure associated with the EryDex suggests that this method of administration may be associated with beneficial effects without the risk of steroid side effects (D'Ascenzo et al, 1997; Crinelli et al, 2000).

7.1.2 *Non-Clinical Trials with EryDex System*

Pharmacokinetics (PK) is the only characteristic of the drug product (DSP) pharmacology that differs from that obtained with the ordinary routes of administration (e.g., oral, IV, intramuscular) due to the novel method of administration by ex vivo encapsulation of the drug into human autologous RBCs that are then infused. The PK of the drug product in humans was evaluated in clinical trials. It is not possible to trial the PK in animal models because the dephosphorylation rate of the pro-drug, DSP, to the diffusible active drug, dexamethasone, in human RBCs is very different from RBC dephosphorylation rates in other species (Zocchi et al, 1991).

7.1.3 *Summary of Human Clinical Trials with EryDex System*

At the time of the IEDAT-04-2022 Protocol finalization, [redacted] participants had been treated with at least 1 dose of EryDex: [redacted] patients in Investigator-initiated studies [redacted] and [redacted] patients in Sponsor-initiated studies (indications: [redacted] with an additional [redacted] healthy volunteer participants who received a single infusion). In addition, 10 patients originally enrolled in the IEDAT-Ery01-2010 trial switched to the Compassionate Treatment Trial. A total of 104 patients with A-T who initially received treatment in Trial IEDAT-02-2015 (ATTeST) continued receiving treatment under Trial IEDAT-03-2018. Continuation of the treatment has then been granted by means of an Expanded Access/Compassionate Use Program, under certain conditions, described in the respective protocols.

Additional detailed information on clinical studies evaluating EryDex (eDSP) can be found in the current version of the EryDex Investigator's Brochure (IB).

Table 2 and Table 3 list the trials performed to date, the indication, the number of participants, the doses administered, the treatment duration, and key safety findings. [redacted]

Table 2: Summary of Clinical Studies of EryDex Conducted to Date for Indications Other Than Ataxia Telangiectasia

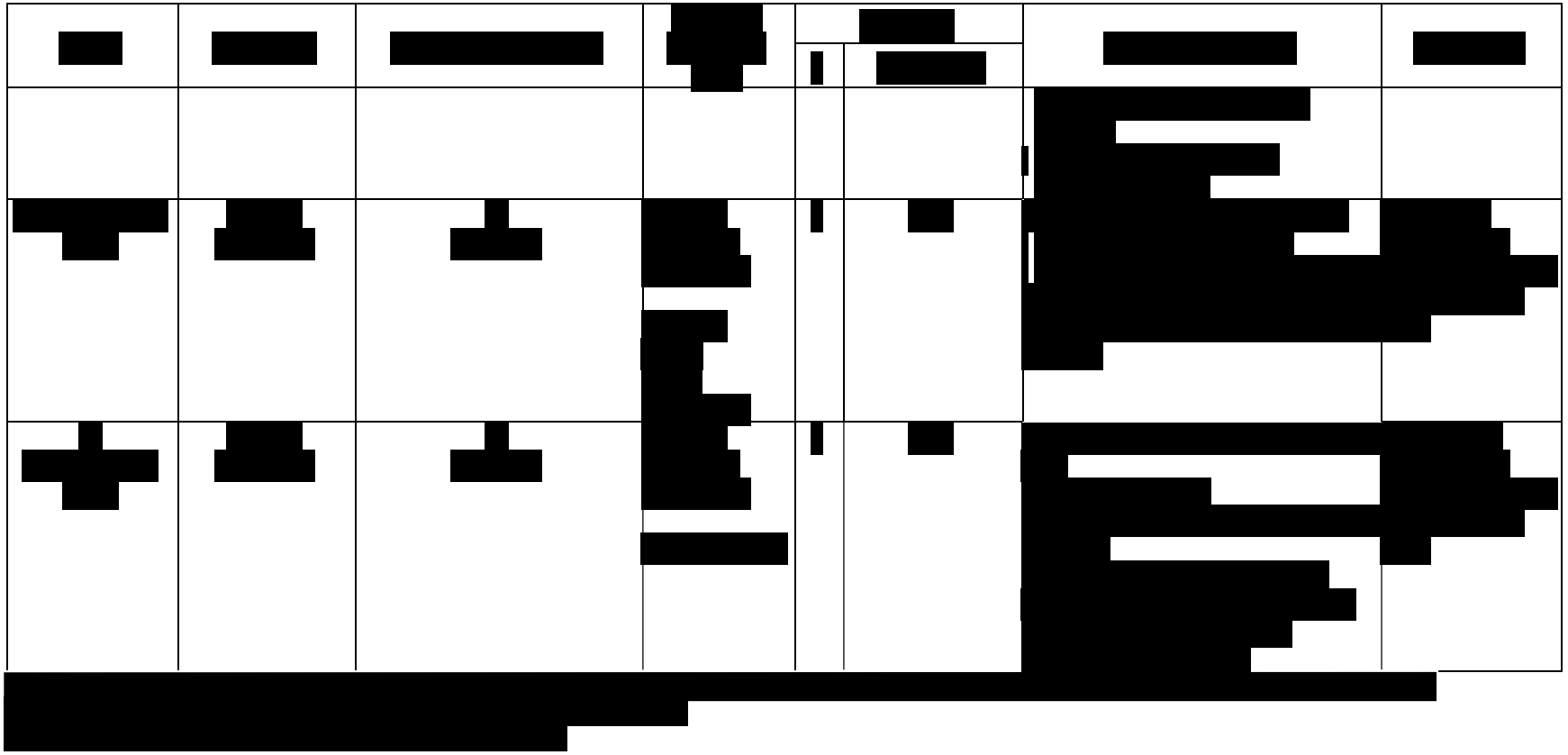


Table 3: Summary of Clinical Studies of EryDex Conducted to Date in the Ataxia-Telangiectasia Population

EryDel Trial #	Indi-cation	#Unique participants	Range of doses (mg DSP)	Infusions		Key safety findings	References
				#	Frequency		
Sponsor-initiated Studies							
1 (IEDAT-ERY01-2010)	AT	EryDex: 22	[redacted]	[redacted]	[redacted]	Premature terminations: 4 (18%) - Withdrawal of consent (1); - AE (lab abnormality - 2); - Protocol violation (1) SAEs: 2 (9%); pneumonia and bronchiectasis; both considered not related to trial treatment; Most frequent AEs (>10%): Otitis (3), Cough (3), Fever (3), Flu syndrome (3)	IEDAT-ERY01-2010 Clinical Trial Report (Data on file: EryDel SpA)
2 (IEDAT-02-2015)	AT	175	[redacted]	[redacted]	[redacted]	Premature terminations before Month 6 visit: 43 (24%) - AE (6) - Withdrawal of consent (11) - Physician decision (1) - Other: • Covid-Related Treatment/Visit Delay (25) Premature terminations after M6 and before the end of the trial: 25 (24%) - AE (1) - Withdrawal of consent (3) - Physician decision (0) - Other • Covid-Related Treatment/Visit Delay (20) • Other reason: 1 41 SAEs. During the entire treatment period, 13.6%, 15.8%, 21.1% of treated participants experienced an SAE, respectively in EryDex Low Dose, High Dose, and Non-switch placebo.	IEDAT-02-2015 Clinical Trial Report

EryDel Trial #	Indi-cation	#Unique participants	Range of doses (mg DSP)	Infusions		Key safety findings	References
				#	Frequency		
3 (IEDAT-03-2018)	AT from Trial IEDAT-02-2015)	104	[redacted]	[redacted]	[redacted]	12 participants (11.5%) experienced 19 SAEs. None of these events were reported as related to the trial treatment (they were all reported as not related, with the exception of 1 "Pneumonia" reported as unlikely related)	IEDAT-03-2018 Clinical Trial Report

Abbreviations: AE = adverse event; AT = ataxia- telangiectasia; COVID = coronavirus disease 2019; DSP = dexamethasone sodium phosphate; SAE = serious adverse event; SD = standard deviation. Summaries of studies conducted to date in healthy volunteers and in the A-T population are provided in Sections 7.1.3.1 through 7.1.3.5.

7.1.3.1 Clinical Study IEDAT-ERY01-2010: Pilot Proof of Concept Phase 2 Trial in Patients with Ataxia Telangiectasia [Chessa et al., 2014]

Title: Evaluation of Effects of Intra-Erythrocyte Dexamethasone Sodium Phosphate on Neurological Symptoms in Ataxia-Telangiectasia Patients

This was a single-arm, open-label, 6-month, Phase 2 trial to assess the effect of EryDex (DSP encapsulated in autologous erythrocytes by the EDS process) on neurological symptoms of patients >3 years of age with A-T enrolled in 2 centers in Italy.

The primary objective of the trial was to evaluate the effect of EryDex in improving neurological symptoms of A-T patients over a 6-month treatment period, assessed by means of the ICARS.

All participants were treated with 6 monthly infusions of EryDex with a target dose of ~10-15 mg of DSP with a one-month period between doses. A total of 26 patients with A-T were screened and 22 were enrolled in the trial, representing the Intent-to-treat (ITT) population. Four participants discontinued prematurely; one withdrew consent, another participant was dropped due to a protocol violation (CD+4 lymphocytes below cut-off at baseline), and 2 participants experienced laboratory abnormalities (decrease in CD4+ lymphocytes count) that led to their withdrawal.

Sixteen participants (72.7%) received all 6 infusions of EryDex as planned. Results for the primary efficacy measure (change from total baseline ICARS score) for the ITT population (n = 22) indicated an overall statistically significant (p=0.02 repeated-measures analysis of variance) improvement with EryDex treatment. Significant improvement was observed after 3 and 6 months of treatment with EryDex for the kinetic sub-score of the ICARS, the clinician rated the Investigator Global Assessment, adaptive behavior (as assessed by the Vineland Adaptive Behavior Scales [VABS]) and ocular motility.

The safety population included all 22 participants. Overall, 15 (68.2%) of participants experienced a total of 29 treatment-emergent adverse events (TEAEs, most of which were rated as mild (~60% of participants) and not related to the trial medication (>90% of participants), with the exception of 1 participant with a mild hypercholesterolemia. Two serious adverse events (SAEs) were reported during the trial, including one case of severe pneumonia and a second participant with bronchopneumonia, and bronchiectasis and bleeding; both participants required hospitalization. Both cases were considered unrelated to trial medication and resolved with treatment. Two participants had a >20% decrease in CD4+ lymphocytes count during the trial period which resulted in premature discontinuation and were considered related to trial medication.

There were no clinically meaningful changes in mean values for laboratory parameters with EryDex treatment, except for:

- serum iron showed a >20% mean decrease from baseline value, with 8 participants having newly occurring abnormal (low) values at the final visit (one reported as a TEAE)
- urinary cortisol showed an approximately 30% decrease from baseline to the final visit (statistically significant, p=0.016). Although this could indicate an effect of EryDex on the hypothalamo-pituitary-adrenal axis, a comparison of blood and urinary cortisol levels versus dose did not indicate any relationship.

No clinically significant changes with EryDex treatment were observed for vital signs, electrocardiograms (ECGs), or physical examination findings.

The level of DSP loading indicated variability in RBC encapsulation across participants. Almost half of the participants had mean DSP dose of less than 5 mg, whereas a DSP dose between 10-15 mg was targeted.

The variability was caused by sub-optimal encapsulation conditions associated with the EryDex process used in this trial. Pilot observation that higher DSP load appears to be associated with improved response of the ICARS led the Sponsor to target higher DSP doses in future efficacy studies.

7.1.3.2 Clinical Study IED-PK01-2013

Title: Pharmacokinetic Study to Measure Plasma Concentrations of Dexamethasone following EryDex (Dexamethasone Sodium Phosphate Encapsulated in Autologous Erythrocytes) Infusion in Healthy Volunteers.

This Phase 1, open-label, single-center (United States [US]), uncontrolled trial was performed to characterize the PK properties of 2 planned doses [redacted] of EryDex using a new version of the EDS process (version 3.2.0). The trial also evaluated the safety and tolerability of the 2 different dose ranges based on TEAEs, SAEs, laboratory parameters, vital signs, ECGs and physical/ neurological examination findings, with the safety of the lower dose (Group 1) being assessed before proceeding to the higher dose (Group 2).

[redacted]

There were no SAEs, or discontinuations due to adverse events (AEs) in either group. Five of 9 (55.6%) participants in Group 1 reported a total of 5 TEAEs, all of mild intensity. In Group 2, 5 participants (55.6%) reported a total of 6 TEAEs, 4 of mild and 3 of moderate intensity. All TEAEs resolved without any sequelae. A number of small but statistically significant transient changes in hematological and biochemical parameters associated with DSP administration were observed. Sporadic abnormal values (high/low) were noted for some of the laboratory hematology and biochemistry parameters at Day 2 and/or the final visit (Day 42); however, none of these were considered to be clinically significant. No meaningful changes from baseline were observed in any urinalysis parameter throughout the trial. [redacted]

[redacted]

7.1.3.3 Clinical Study Ery51Cr-01-2014

Title: Determination of the In Vivo Recovery and Survival of EryDex (Dexamethasone Sodium Phosphate Encapsulated in Autologous Erythrocytes) in Non-patient Volunteer Subjects.

This was a randomized, single-blind, single-center, concurrently controlled, exploratory, Phase 1 trial to determine the in vivo kinetics (24-hour post-infusion recovery and time to disappearance of 50% of the labeled RBCs from the circulation [T50] survival) of EDS-processed autologous RBCs in non-patient volunteers. [redacted] Safety assessments included monitoring for AEs, routine laboratory tests (hematology, biochemistry, urinalysis), vital signs, physical examinations, ECG evaluations, serum pregnancy testing for fecund women, and documentation of concomitant medications.

[redacted]

The results for the mean RBC recovery for the Active Treatment Arm indicate that DSP-loaded EDS-processed cells met the FDA criteria for 24-hour RBC recovery ≥75% (Dumont and AuBuchon, 2008). [redacted]

[redacted]

There were no discontinuations due to AEs, SAEs or any AEs rated severe or unexpected. There were no clinically notable changes detected in measures evaluating a pre-specified hemolysis panel of tests, or other routine laboratory hematology, biochemistry, and urinalysis parameters. Similarly, there was no evidence

of a systematic effect on vital signs, weight, or ECGs, and no pattern of AEs was detected with the trial treatments.

7.1.3.4 Clinical Study IEDAT-02-2015: Phase 3 Trial of Ataxia-Telangiectasia Treatment with EryDex SysTem - ATTeST

Title: A Multi-center, Randomized, Double-blind, Placebo-controlled Trial to Evaluate the Effects of Intra-Erythrocyte Dexamethasone Sodium Phosphate on Neurological Symptoms in Patients with Ataxia-Telangiectasia

The ATTeST trial was a multi-national, multi-center, 12-month, randomized, double-blind, placebo-controlled, Phase 3 trial, designed to assess the effect of 2 non-overlapping dose ranges of EDS (DSP in autologous erythrocytes), administered by IV infusion once per month, on neurological symptoms of patients with A-T. The results for the ATTeST trial formed much of the basis for the design of the IEDAT-04-2022 trial and are described in some detail.

Patients were randomly assigned to one of the 3 treatment groups:

- EryDex Low Dose [redacted] infusion
- EryDex High Dose [redacted] infusion
- Placebo EDS infusion

Participants who completed the 6-month Initial Treatment Period were eligible to continue treatment for an additional 6 months in the Extension Treatment Period. Participants originally randomized to one of the 2 dose ranges of EDS (Groups 1 or 2) continued the same treatment. Those originally randomized to the Placebo group (Group 3) were re-allocated in equal proportions (1:1) and received either the EryDex Low Dose or High Dose (one third at 6 months and one third at 9 months). Participants who completed the last trial visit were eligible to enroll in an open-label extension (OLE) trial (IDEAT-03-2018).

Males and females, at least 6 years of age, with neurological signs of A-T (incoordination of the head and eyes in lateral gaze deflection and gait ataxia associated with an inappropriately narrow base), an autonomous gait (or helped by support), and a genetic diagnosis of A-T (at least 1 Ataxia Telangiectasia Mutated [ATM]) and/or ATM protein deficiency by Western blot, were included. Patients with low CD4+ lymphocyte counts, current/previous neoplastic disease, history of severe impairment of the immunological system, chronic conditions representing a contraindication to the use of steroid drugs, or any other significant disease or concomitant medication that could have put the patient at risk were excluded. Females of childbearing potential that were pregnant, breastfeeding or not using adequate contraception were also excluded. Eligible patients could not have participated in a trial with another investigational agent within 30 days of the start of the Screening Period and must have discontinued prior steroid medication at least 4 weeks before the first dose of EryDex.

The primary efficacy objective for the initial 6-month treatment period was to evaluate the effect of 2 dose ranges (Low Dose and High Dose DSP/infusion) of EryDex, compared to placebo, on CNS symptoms measured by the change in mICARS from baseline to Visit 9 (Month 6) in participants with A-T.

EryDex was also compared to placebo using the Clinical Global Impression of Change (CGI-C) on neurological symptoms of A-T from baseline to Month 6 (Visit 9).

Other secondary efficacy objectives included:

- Evaluate the effect of EryDex, compared to placebo, on the Clinical Global Impression of Severity (CGI-S; structured) of neurological symptoms of A-T from baseline to Month 6 (Visit 9).

- Evaluate the effect of EryDex, compared to placebo, on adaptive behavior measures by the VABS from baseline to follow-up based on repeated measures at Month 3 (Visit 6) and Month 6 (Visit 9).

Sub-group analyses by age (younger [6 to <10 years] and older [≥10 years]) were conducted, and randomization procedures ensured balance across treatment groups. The importance of this age-based sub-group is supported by natural history data (Rothblum-Oviatt et al., 2016; Jackson et al., 2016) and clinicians' observations, which show that the neurological deterioration in patients with A-T follows a pattern over time where the majority of the clinical signs of the neurodegeneration are observed before age 10. By the age of 12, the vast majority of A-T patients become wheelchair-dependent and the neurological signs of disease progression slow significantly primarily affecting upper limb and respiratory functions.

ATTeST Efficacy Results

Among the 176 participants randomized, efficacy analyses were performed on 164 participants (Full Analysis Set [FAS] population) who received at least one dose of trial treatment and who had the trial primary outcome assessed at the month 6 evaluation. Of the 164 participants in the FAS, 54 were randomized to placebo, 56 to EryDex low dose, and 54 to EryDex high dose.

As noted above, the trial utilized ICARS for efficacy measures (Trouillas et al., 1997), which is a 100-point semi-quantitative scale offering a compartmentalized quantification of the following 4 sub-scores: Posture and gait disturbances, kinetic functions, speech disorders, and oculomotor disorders. The change in mICARS from baseline to Month 6 (Visit 9) was evaluated as the primary endpoint, per the Statistical Analysis Plan (SAP).

This analysis was also performed using a Rescored Modified International Cooperative Ataxia Rating Scale (RmICARS), and on the ICARS, according to the SAP. Please refer to Table 4 for a comparison of the ICARS, mICARS and RmICARS. The primary endpoint analyses were performed based on readings performed by blinded central reading process, as specified in the protocol. Blinded local readings were also performed and available for analyses. Information gained during ATTeST using both methods, central and local scoring informed the decision to use local blinded readings in IEDAT-04-2022 trial; please refer to Section 7.7 for further details.

Results from the ATTeST trial clearly showed that, compared to older participants, EryDex had a much larger treatment effect both for the primary and key secondary endpoints, in the 6- to 9-year-old subpopulation.

[redacted]

The analysis of the key secondary efficacy measure (CGI-C at Month 6) supports the findings for the primary endpoint. [redacted]

In the subgroup analysis of participants based on age, the proportion of 6- to 9-year-old participants in the high-dose EryDex group who showed improvement (CGI-C scores between 1-3) [redacted]

[redacted]

Other secondary efficacy analysis included CGI-S score and VABS score. [redacted]

ATTeST Safety Results

The safety objective was to evaluate the safety and tolerability of EryDex, compared to placebo, in A-T participants, based on the occurrence of TEAEs, including SAEs and discontinuations due to AEs, and changes in vital signs, laboratory parameters, ECGs, and physical/neurological examination findings over the 12 months total double-blind trial duration.

Of the [redacted] participants who were randomized, [redacted] participants received trial treatment and were included in the Safety Population (SP). [redacted] respectively, for those who remained in their initial treatment groups.

The safety data were analyzed with particular attention to overall TEAEs, potentially steroid-related AEs including increased risk of infection, and bone mineral density (BMD), and Tanner scale as indicators of long-term adverse effects of chronic steroid treatment. The overall conclusion is that these data do not raise significant clinical concerns to the overall safety of the EryDex treatment, nor concern regarding the known long-term side effects of chronic steroid treatment, consistent with the Independent Data Safety Monitoring Board (iDSMB) regular reviews throughout this trial and the ongoing OLE-IEDAT (IEDAT-03-2018) trial.

EryDex treatment was well tolerated, with only mild to moderate transient AEs that rapidly resolved in any dose group. No pattern of clinically relevant AEs was observed including steroid-related side effects. Serious adverse events with an incidence of 5% or higher were reported by 8 (13.6%) patients in the low-dose, 9 (15.8%) participants in the high dose, and 4 (21.1%) participants in the placebo groups through the end of the Extension Treatment Period.

The most commonly occurring SAE in the low-dose, high-dose, and placebo groups, was bacterial test positive for samples of the final bag before infusion in the initial treatment period, (5.1%, 8.8%, and 6.8%, respectively) as well as through 12 months of Extension treatment period (6.8%, 10.5% and 15.8%). A root cause analysis was performed and showed that these positive tests were a result of external contaminations during the handling process of sampling for bacterial testing and demonstrated that no participant had a positive blood culture post infusion, and no participant experienced any clinical or laboratory symptoms of bacteremia. Subsequently, following [redacted] further cases of "external" contaminations were reported in the regular safety update reports (thus, not as SAEs).

The effect on BMD was assessed by BMD Z-score. Based on the mean BMD Z-scores, adverse effects on BMD due to EryDex treatment were not observed (a Z-score of < -2.5 means osteoporosis, between -2.5 and -1.0 osteopenia, and above -1 normal; a decrease of <0.5 has no clinical meaningfulness). Most patients stayed at their baseline category for the 12-month ATTeST treatment period, and very few went from one category to another during that timeframe. For the high-dose EryDex group, only 7 patients within the 12-month trial moved from normal to abnormal (not clinically significant) and 1 patient from

normal to abnormal (clinically significant), while 1 patient returned from abnormal back to normal (not clinically significant).

Laboratory parameters of special interest included alpha fetoprotein, CRP, and haemoglobin A1c. Alpha-fetoprotein is pathologically elevated in most A-T patients, and CRP is a marker of inflammation, which could be associated with steroid treatment. None of the laboratory parameters of special interest showed a possible negative influence of steroid treatment over the entire trial.

Effect on development was assessed using the Tanner scale. Numbers and percentages do not indicate any unusual pattern that would indicate an influence of EryDex treatment on the normal child and adolescent development of the trial population (Stage I is initial puberty and Stage V is completed puberty).

Suicidal ideation severity of observed cases was assessed monthly throughout the ATTeST trial, by administering the Columbia Suicidal Severity Rating Scale (C-SSRS). Severity scores ranged from 0 (no ideation present) to 5 (active ideation with plan and intent). During the initial treatment period, 3 baseline cases (2 cases with severity score 1 and 1 case with severity score 4) were only observed in the placebo group. At month 1, 1 case was observed in placebo (severity score=1) and 1 case in high dose (severity score=2). At month 3, 1 case was observed in the high-dose group (severity score=1). At months 7 and 10, 1 case (severity score = 1) each was observed in a participant who switched from placebo to high dose.

7.1.3.5 Clinical Study IEDAT-03-2018: Open-label, Long-term, Extension Treatment using Intra-Erythrocyte Dexamethasone Sodium Phosphate in Patients with Ataxia Telangiectasia Who Participated in the IEDAT-02-2015 Study (OLE-IEDAT)

This was an international (North America, Europe, Africa, Asia and Australia), multi-center, prospective, open-label treatment trial, designed to continue to provide long-term monthly EryDex treatment to participants, regardless of assigned treatment group, who participated in the IEDAT-02-2015 trial.

The trial aimed to collect information on the long-term safety and efficacy of the trial treatment. Participants received monthly infusions of EryDex (with a dose correspondent to the ATTeST high dose). If this dose of EryDex was not tolerated, a participant was discontinued from the trial. During the trial, long-term efficacy assessments were performed approximately every 6 months, while safety parameters were assessed at each monthly visit. [redacted]ts have been enrolled.

Below is a summary of safety data, based on the outputs generated in September 2021, following an interim data cut (final database lock has not occurred at the time of the finalization of the IEDAT-04-2022 trial protocol).

Safety data are available for [redacted] participants in the trial, who were followed up for as long as [redacted] There were no deaths. A total of 12 participants experienced a total of 19 SAEs. The SAEs reported included: B-Cell lymphoma, Hodgkin's disease, pneumonia, dysphagia, odontogenic cyst, thrombocytopenic purpura (×5), pyrexia (×2), forearm fracture (×2), viral infection, lower respiratory tract infection, central nervous system lesion, gastrointestinal tube insertion, and pharyngeal swelling. None of these were reported as related to trial treatment (i.e., all reported as not related, except for "Pneumonia" reported as unlikely related) and all were reported as resolved. Three participants had a total of 4 AEs that were reported as leading to permanent discontinuation of trial treatment. One participant had the events of B-Cell lymphoma Hodgkin's disease (see SAE above) and decreased appetite, both of which were noted as leading to discontinuation of trial treatment. The SAE of B-Cell lymphoma Hodgkin's disease, noted above, and an AE of mood altered were also noted as leading to disconnection of trial treatment. The AE of mood altered was categorized as probably related to trial treatment.

Overall, at least 1 TEAE was noted in 94.2% of participants, with 36.5% having at least 1 event categorized as at least possibly related to trial treatment. The TEAEs reported in at least 5% of participants included: pyrexia (31.7%), nasopharyngitis (20.2%), vomiting (19.2%), diarrhea (22.1%), pruritus (6.7%), iron deficiency (15.4%), upper respiratory infection (22.1%), cough (18.3%), fatigue (15.4%), headache (14.4%), erythema (6.7%), bronchitis (9.6%), infusion related reaction (26.9%), low ferritin (7.7%), skin papilloma (7.7%), rash (8.7%), and anemia (5.8%), coronavirus infection (16.3%), urinary tract infection (5.8%), fall (6.7%), nausea (6.7%), rhinorrhea (11.5%), epistaxis (6.7%), Coronavirus test positive (6.7%), blood LDH increased (5.8%), nervous system disorder (28.8%), metabolism and nutrition disorder (26.9%), musculoskeletal and connective tissue disorder (21.2%), product contamination (12.5%), eye disorder (10.6%), psychiatric disorders (10.6%), vascular disorder (9.6%), and renal and urinary disorder (6.7%).

Only 3 (2.9%) participants were reported as having osteoporosis, and 4 participants (3.8%) had osteopenia. At Baseline, 81 participants had BMD assessed and 11 of them presented abnormal clinically significant bone density. Nine participants with abnormal clinically significant bone density shifted from abnormal clinically significant to abnormal not clinically significant during the first 12 months of trial. The analysis of the shifts from Baseline also showed that at each post Baseline timepoint, a maximum of 3 occurrences of shift to abnormal clinically significant was reported; on the contrary, at several timepoints no shifts to abnormal clinically significant results were reported.

Overall, these findings during long-term follow-up of treated participants are consistent with expectations for the trial populations as being part of the disease progression and do not raise new safety concerns.

All participants who did not discontinue the OLE-IEDAT trial and who consented to participate have been transitioned to an Expanded Access/Compassionate Use Program at the time of the finalization of this protocol, with more than 50 participants already in treatment in Expanded Access/Compassionate Use Program.

7.2 Drug Product and Delivery System

7.2.1 *Generic Name*

Drug: Dexamethasone sodium phosphate solution (25 mg/mL), referred to as DSP Solution

Medical Devices: Blood processing equipment and kit of single-use sterile disposable device accessories: Hypertonic/hypotonic sterile solutions (Process Solutions); Loader electromedical equipment.

7.2.2 *Proprietary Name*

Drug: Not determined (a proprietary name for DSP solution, 25 mg/mL has not yet been proposed)

Medical Devices: EryKit_01; Syringe Kit; PIGPA Hypertonic Solution, Hypotonic Solution 1, and Hypotonic Solution 2 (Process Solutions); Red Cell Loader (RCL).

7.2.3 *The EryDex System*

The EryDex system is used to load DSP to autologous erythrocytes [redacted]

[redacted]

In the European Union (EU), the RCL, the EryKit_01, the Syringe Kit, and process solutions are medical devices in compliance with the EC Council Directive MDD 93/42/EEC of 14 June 1993 and following amendments. The RCL, EryKit_01, Syringe Kit, and process solutions have been Conformité Européenne (European Conformity; CE) marked in the EU as Class IIb medical devices since July 2010 in accordance with the requirements of MDD 93/42/EEC Annex II. The RCL, EryKit_01 and Syringe Kit are also in compliance with the Medical Device Regulation (EU) 2017/745 of 05 April 2017 and following amendments, while the process solutions are in the transition phase. In the US, the RCL, the EryKit_01, the Syringe Kit, and the process solutions have been classified as medical devices and used in clinical studies under an IND [redacted]erapeutics S.p.A is the manufacturer of the RCL, EryKit_01, Syringe Kit, and process solutions.

Additional details of the EDS components and the process for encapsulating DSP in human erythrocytes can be found in Appendix 2.

7.2.3.1 EryDex Process Training and Documentation

All site personnel involved in blood collection, processing, testing, and administration of EryDex will be trained on the use of the following checklist (will be provided as a separate trial document), to be completed at each treatment visit:

- The EryDex checklist: to collect information on the EryDex process, used materials (including labels from the infusion bag and randomized treatment ampule), and infusion.
- "Aseptic Procedure Guideline", a stand-alone document that includes detailed description of the actions to be implemented to avoid contaminations in the following steps:
  1. Aseptic procedures for prevention of contamination of EryDex
     - General background information
     - Phase 1: Collection of [redacted] autologous blood cells
     - Phase 2: EryDex Process
     - Phase 3: EryDex administration
  2. Technical instructions for EryDex sterility sampling
     - Samples for sterility culture test, CBC and determination of DSP content

In addition, the EryDex Operators will be trained and receive copies of the following manuals:

- Operator's Manual EryDex System: contains all information required for the use of the RCL and EDS, including:
  - Safety information and warning
  - Product description and technical specifications
  - Instructions for installation of RCL
  - Description of each step of the EryDex process (gather supplies, setup, blood collection, blood connection, EryDex process, EryDex infusion, removal and disposal)
  - Maintenance instructions and after-sales service
- Quick Reference Guide EryDex System: contains a brief guidance related to the use of the EDS including:
  - EryDex process warnings
  - Identification of the parts of the RCL
  - Identification of the parts of the EryKit_01
  - Needed supplies
  - Tips related to setup, blood collection, blood connection, EryDex process, EDS-EP infusion, removal, and disposal.
- Instructions for Use of EryKit_01: contains all information required for the use of EryKit_01, Hypotonic Solutions 1 and 2, PIGPA Hypertonic Solution within the EDS, including:
  - Safety information and warning
  - Product description and technical specifications
- Instructions for Use of Syringe Kit: it contains all information required for the use of Syringe Kit within the EDS, including:
  - Safety information and warning
  - Product description and technical specifications
  - Product use

7.3 Risk/Benefit Assessment

7.3.1 *Risks*

Dexamethasone is a steroid class drug first approved by FDA in 1958, therefore there is a large body of information known about its risk. It is used in the treatment of a number of chronic (long-term) inflammatory diseases. Known risks of dexamethasone treatment include allergic reaction, itching, increased appetite, irritability, insomnia, fluid retention, dyspepsia, nausea, vomiting, muscle weakness, impaired wound healing, hyperglycemia, headaches, dizziness, mood swings, dizziness, blurred vision, muscle cramps, cataracts, hypertension and bone loss. Mild to moderate itching during the EryDex infusion procedure was observed in prior studies.

Participants in the trial will be monitored each month through physical exam, vital signs, laboratory testing and AE surveillance to identify development of dexamethasone-related side effects.

The safety data from the most recent and largest trial of EryDex in the A-T population (IEDAT-02-2015 ATTeST) were analyzed with particular attention to potentially steroid-related AEs including increased risk of infection, bone density, and Tanner scale as indicators of long-term adverse effects of chronic steroid treatment. The overall conclusion, corroborated by the preliminary safety data from the IEDAT-03-2018 (OLE-IEDAT) trial, was that these data did not raise clinical concerns to the overall safety of the EryDex treatment, nor concern regarding the known long-term side effects of chronic steroid treatment, consistent with the independent regular iDSMB reviews throughout the ATTeST and OLE-IEDAT trial.

Risks associated with blood collection for laboratory tests include pain, swelling, bruising and, on rare occasions, infection at the injection site. [redacted]

Risks associated with the EryDex infusion are similar to any blood infusion, and include pain, bruising, infection, hypotension, fever, chills, nausea, hives, rash, itching, swelling of the face or neck, difficulty breathing, and anemia. Pruritus and itching are known side effects of IV administration of steroids, and in prior studies, some patients experienced these side effects. Mitigation methods were implemented in IEDAT-02-2015 and IEDAT-03-2018 studies (i.e., pre-infusion Benadryl and slowing down speed of infusion), which resolved this issue in the majority of cases. It should be noted that the reported itching and pruritus subsided within less than a minute after completing the infusion procedure. These mitigation methods will be employed, as needed, in this trial.

The likelihood of transfusion reactions is very low, since autologous RBCs are being infused. Participants will be monitored throughout the infusion process. Within 15 minutes after the infusion, vital signs assessment will be performed, and the observation period may be extended, based on Investigator's judgement. Physical examination must be conducted on the day of trial treatment administration.

Sterility testing will be performed before each infusion, by means of blood culture test, and after each infusion, using an EryDex sample from the satellite bag. Appropriate actions to be taken, should there be a positive culture, are detailed in Appendix 6.

A bone density test is obtained using dual-energy x-ray absorptiometry. This is a non-invasive, painless imaging trial that uses a very small dose of x-ray to measure the density of bones. The scan involves a very

low amount of radiation exposure—less than a day's exposure to natural radiation, and less than one-tenth of the dose of a standard chest x-ray.

7.3.1.1 Risks During Pregnancy and Lactation

In pregnant rats, dexamethasone crosses the placenta, but fetal plasma levels are below maternal levels. To a small extent, dexamethasone distributes into breast milk. The genotoxicity potential of dexamethasone has been evaluated in in-vitro and in-vivo studies, with both sets of experiments indicating it is devoid of genotoxic potential. Dexamethasone appears to be associated with a teratogenic potential in mice, rats, rabbits and monkeys. Post-natal development studies indicate that it has the potential to alter the development of the immune system, heart, kidney, bone, brain tissue, lipid profiles, and social behavior in rat pups.

In the clinical trials performed to date with eDSP, women who are pregnant or lactating have been excluded. In the clinical studies in healthy subjects and patients with A-T, females of childbearing potential who are not using adequate contraception methods or are pregnant or lactating are excluded.

7.3.2 *Potential Benefits*

Results from the clinical trials conducted to date with EryDex in participants with A-T suggest that EryDex treatment may delay A-T disease neurological progression.

7.4 Trial Rationale

Trial IEDAT-04-2022 is an international, multi-center, randomized, double-blind, placebo- controlled, Phase 3 trial to assess the effect of EryDex (DSP in autologous erythrocytes) administered by IV infusion once every 28 days (window +2 days, -7 days), on neurological symptoms of participants with A-T.

7.4.1 *Rationale for Using the EDS in Treating A-T Patients*

EryDex was granted US orphan drug designation for the treatment of A-T on July 24, 2012 (ODD #12-3732). A-T is considered a rare disease in the literature (Orphanet 2014) and the consortium of European partners. There is no marketed drug approved to treat A-T, and there is no treatment available that slows or stops the progression of A-T patients to early debilitating disability and mortality. Symptomatic, preventive, and supportive management of the key disabilities and symptoms of the disease has provided limited benefit. Immune deficiencies are managed by administration of immunoglobulins, and infections are managed by both preventive and symptomatic use of antibiotics. Developmental deficiencies are managed by physical therapies, education, or targeted treatments (Hoche et al, 2012).

Neurological degeneration is the major contributor to the severe outcome of the disease. Progression of the neurological problems is currently unstoppable.

Anti-Parkinson and antiepileptic drugs provide limited benefit in the management of extrapyramidal symptoms. The serendipitous discovery of the benefit of short-term betamethasone in a patient with A-T (Buoni et al, 2006), and further pilot, uncontrolled, short–term treatment trials in small numbers of patients have suggested that betamethasone at doses of 0.03 mg/kg, but not 0.01 mg/kg, given for a short period has beneficial effects on the neurological impairment of A-T patients (Russo et al, 2009; Broccoletti et al, 2008). Treatment discontinuation in these studies was accompanied by the reappearance of symptoms, suggesting that long- term administration may be required to preserve the benefit.

These early findings with administration of betamethasone prompted the search for steroids that could be given at low plasma concentrations for an extended period of time without the associated steroid-like

effects. The EDS, consisting of DSP encapsulated in autologous erythrocytes, allows the administration of dexamethasone at low plasma concentrations that are associated with approximately 80-85% occupation of the glucocorticoid receptor, similar to the glucocorticoid receptor occupancy that occurs with the betamethasone plasma concentrations reported by Broccoletti et al, 2011. The EDS provides the benefit of low and constant plasma delivery of dexamethasone without the associated steroid side effects, despite long-term use in the pediatric population (Rossi et al, 2001; Lucidi et al, 2006; Annese et al, 2005; Bossa et al, 2008; Castro et al, 2007).

The mechanism of action of glucocorticoids in producing the benefits observed in A-T patients was postulated to occur through regulation of expression of corticosteroid-responsive genes, suppression of inflammatory cytokines, or its effect as an antioxidant (Russo L. et al., 2009). More recently, an effect of dexamethasone on ATM gene splicing in an A-T lymphoblastoid cell line, leading to production of a new active ATM protein variant, has been demonstrated (Menotta M. et al., 2012). This may explain in part the beneficial effect of treatment with glucocorticoid analogues in A-T patients. The results show, for the first time in mammalian cells, a short direct repeat-mediated non-canonical splicing event induced by dexamethasone that leads to the skipping of mutations upstream of nucleotide residue 8450 of ATM Coding DNA Sequence.

The clinical program to assess the efficacy and safety of EryDex for the treatment of A-T has included one Phase 2 trial (IEDAT-ERY01-2010), one Phase 3 trial (IEDAT-02-2015 [ATTeST]), one OLE (IEDAT-03-2018 [OLE-IEDAT]) that is coming to its conclusion at the time of the finalization of the IEDAT-04-2022 trial protocol, as well as 2 Phase 1 studies in healthy volunteers (Ery51Cr-01-2014, IED-PK01-2013). All the participants who did not discontinue the OLE-IEDAT trial are currently (at the time of time of the finalization of the IEDAT_04-2022 trial protocol) receiving the option to continue treatment in an Expanded Access/Compassionate Use Program.

7.4.2 *Rationale for the Participant Population*

The selection criteria for this trial have been based on the cardinal symptoms of A-T, considerations on the natural history of the disease, and the prior experience gained through EryDex studies in this population, particularly the Phase 3 ATTeST trial, whose results were used to inform the design of the IEDAT-04-2022 trial, including the definition of the trial population.

A-T is a rare inherited autosomal recessive, multisystem disorder, characterized by the following: progressive neurologic disease, including cerebellar ataxia and oculomotor apraxia; cutaneous and ocular telangiectasia (90%); increased alpha-feto-protein levels (95%); reduced or absent IgA levels (70%) and ATM protein (98%); increased susceptibility to chronic nasal and pulmonary infections (70%); delayed organ maturation; and increased susceptibility to X-rays and malignancies (Hoche et al., 2012). In countries where marriage between cousins is uncommon, such as in the US, the incidence of A-T is about 1 in 40,000 live births. However, among ethnic groups where cousin-cousin marriages are common, the incidence increases significantly. The carrier frequency is approximately 1 in 100 persons (www.cancer.net/cancer-types/ataxia-telangiectasia). The world-wide prevalence of A-T is estimated to be between 1 in 40,000 and 1 in 100,000 live births (Rothblum-Oviatt et al., 2016). A-T is due to mutations in the ATM gene located on human chromosome 11 (11q22.3). These defects in the ATM gene reduce or eliminate the function of the ATM protein that controls cell division and is involved in double-strand DNA repair (Lee and McKinnon, 2000; Boehrs et al., 2007), thus leading to repeated infections and malignancies.

The onset of symptoms becomes evident in early childhood when children demonstrate unsteadiness while walking or swaying when standing still or sitting. Most children with A-T have stable neurologic symptoms for the first 4 to 5 years of life but begin to show increasing problems in early school years. Oculomotor apraxia, slurred speech, and difficulties in swallowing appear in late pre-school and early school age

children. A-T patients are unable to control their muscles by the time they are 10 to 12 years of age, and this may lead to problems with fine motor functions (writing, coloring, and using utensils to eat) and slurring of speech (dysarthria) that usually stop progressing after the age of about 12 to 15 years. Most A-T patients are restricted to wheelchair use for ambulation, with a recent systematic review of the literature finding a mean age of wheelchair requirement at age 10 (Petley, 2022). Involuntary movements such as tremors, myoclonic jerks, dystonia, chorea, and athetosis are variable in the age of onset and rate of progression (Boder, 1985; Perlman et al, 2003; Chun and Gatti, 2004). The immune system becomes progressively weaker, and recurrent respiratory infections and blood cancers are noted in late-stage patients. There is high variability in life-expectancy; however, most patients die around the age of 25 years, largely due to chronic lung disease or malignancies (Poupard, 2003; Crawford et al, 2006).

Approximately 86 participants between 6 and 9 years old will be enrolled and analyzed as primary analysis population; approximately 20 participants ≥10 years old will also be recruited.

In the ATTeST trial, there was a much larger EryDex treatment effect in the 6- to 9-year-old subpopulation. These findings are aligned with the natural history data that show a slow rate of onset for A-T symptoms up to age 4-5 years, increasing from age 6 to 9 years and then decreasing significantly from age 10 years onwards.

Based on the natural history data and results from ATTeST, the selection of the 6- to 9-year-old population for this Phase 3 trial may allow for clear separation from placebo and demonstration of a treatment effect over a 6-month treatment, in which these participants show a relatively fast deterioration of their neurological symptoms.

The lower age limit of 6 years ensures that participants can provide assent in most of the cases. In addition, children with A-T have limited clinical signs of neurological deterioration until about 4 to 6 years of age (Rothblum-Oviatt et al., 2016; Jackson et al., 2016). The onset of neurological symptoms is rare before 6 years, and any worsening that occurs is potentially offset by the improvement due to age-associated development. Furthermore, most clinicians indicated children below age 6 are largely unable to comply with the physicians' requests during the administration of the ICARS, which is a scale that requires great cooperation in the completion of the different tasks.

Finally, according to the guidelines published by the World Health Organization (Howie, 2011), relevant to safe limits in blood withdrawal in paediatric participants, there are restrictions on the total amount of blood that can be withdrawn according to total blood volume, and therefore to body weight, over time. Based on such guidelines, participants below 15 kg of weight cannot undergo a blood withdrawal of 50 mL within 24 hours. A-T children typically fall at or below the 3rd centile on the CDC charts (Natale et al., 2021), with a rate of poor growth that varies based on several factors, including the severity of the disease. Therefore, taking into account the possible variability in growth, A-T children below 15 kg are likely to be younger than 6 years of age.

The trial will include males and females, with genetic confirmation and neurological signs of A-T (incoordination of the head and eyes in lateral gaze deflection, gait ataxia associated with an inappropriately narrow base). Participants must have an autonomous gait or be helped by a support (i.e., ICARS score for *Item 1 – Walking Capacities* between 0 and 4, included).

Additionally, participants must have a body weight ≥15 kg, and CD4+ lymphocytes count of >400/mm^3 (for participants less than 7 years old) or >150/mm^3 (for participants ≥7 years old). In the presence of oral infections, like oral candidiasis, documented at the screening or recurrent as per medical history documentation, the limit increases to >200/mm^3 (for participants ≥7 years old). The CD4+ lymphocyte counts have been included to assess the immune status of these participants, as the ATM gene compromises their ability to combat infections. CD4+ lymphocyte cut-off limits are stratified by age as recommended by

A-T experts. If participants have CD4+ lymphocyte counts consistently below 150/mm^3 (for participants ≥7 years old), or below 200/mm^3 (for participants ≥7 years old and in the presence of oral infections, like oral candidiasis, documented at the screening or recurrent as per medical history/AEs documentation) or below 400/mm^3 (for participants less than 7 years old) during treatment, the Principal Investigator (PI) should contact the Medical Monitor to discuss if it may be necessary to interrupt trial treatment.

The exclusion criteria for the trial are designed to exclude participants who would be at increased risk for an adverse outcome related to treatment with EryDex or their participation in the trial due to their medical history/current status or use of concomitant medication.

Potential participants failing the ACTH stimulation test will be excluded from the trial and referred to an endocrinologist.

Dexamethasone has teratogenic potential in rodents; therefore, any females who are pregnant or breastfeeding will be excluded from participation. Female participants will have a serum pregnancy test obtained at Screening. Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated before each infusion at Visits 1, 4, 5, 6, 7, 8 and a serum pregnancy test at Visit 9/Early Discontinuation if noted to be at Tanner Stage 2 or greater at this Visit. Females of childbearing potential should use an adequate birth control method in order to be eligible. For further details on the adequate contraceptive measures, please refer to Section 14.7.

Patients with A-T are at increased risk of neoplastic and immunological diseases due to the decreased ability of the ATM gene to repair DNA; therefore, patients with these conditions are excluded. Patients who are currently on steroids, or have taken them within 6 weeks before Baseline, are excluded, as their effects may confound the assessment of the efficacy and safety of EryDex.

7.5 Rationale for the Trial Design

7.5.1 *Double-blind Design*

The double-blind design was chosen to reduce the potential for bias in efficacy and safety assessments, since neither the Investigator, the participant, nor Sponsor/Contract Research Organization (CRO) know the treatment assignment. The double-blind design also minimizes bias in assessing the potential relationship of safety findings to the trial treatments.

7.5.2 *Use of Placebo control*

The trial will evaluate eDSP compared to placebo in the treatment of participants 6 to 9 years old with A-T. Currently, there is no approved treatment for A-T; therefore, no active control has been included in the trial. The placebo control will allow testing of the null hypothesis and provide a statistically valid assessment of the effects of eDSP treatment.

7.5.3 *Stratification of Randomization by Sex and Region*

To decrease potential heterogeneity between treatment groups in baseline symptomatology related to differences between groups, the randomization will be stratified by age (6 to 9 years old and ≥10 years old), sex (male, female), and by geographic region.

7.5.4 *Trial Treatment Duration*

The 6-month, placebo-controlled comparison is considered adequate to assess the symptomatic effects of eDSP on most motor disorders, including ataxias.

7.5.5 *Collection of race/ethnicity*

The participants' race and ethnicity will be collected because analysis of results according to race/ethnicity are required by several Regulatory Authorities (e.g., black population for the Food and Drug Administration [FDA] in the US).

7.6 Rationale for the eDSP Dose

The eDSP dose that will be administered in the IEDAT-04-2022 trial corresponds to the high dose of IEDAT-02-2015 (ATTeST) trial, which is obtained [redacted]

Dose selection for trial IEDAT-04-2022 is based on the analysis of the efficacy and safety results of ATTeST. A dose response was observed favoring the high dose and analyses of the 6–9-year-old population for RmICARS and CGI-C suggested superiority of the high dose.

In the combined safety analysis of ATTeST and the open-label extension trial, EryDex high dose was well tolerated, with no differences versus placebo or EryDex low dose.

Based on these data showing that a higher dose was shown to correlate with a greater response in the ATTeST, and safety data suggesting there is no clinically meaningful difference in the safety profile of the 2 doses, the same dose will be used in the IEDAT-04-2022 trial.

For an individual participant, if this dose shows significant tolerability issues that do not resolve, the participant will be discontinued from the trial treatment and appropriate medical follow-up will be performed.

7.7 Rationale for Efficacy Measures

The efficacy measures for this trial were selected based on feedback from Regulatory Authorities and data from the completed ATTeST trial. The efficacy measures to be used in the trial are described below.

7.7.1 *Rescored modified International Cooperative Ataxia Rating Scale (RmICARS)*

The ICARS is a 100-point semi-quantitative scale offering a compartmentalized quantification of the following 4 sub-scores: Posture and gait disturbances, kinetic functions, speech disorders, and oculomotor disorders. The internal consistency, criterion-related validity, and internal construct validity of the ICARS have been established in patients with focal cerebellar lesions (Schoch B et al, 2007). The inter-rater reliability, test-retest reliability and internal consistency have also been validated in patients with spinocerebellar ataxia (Weyer et al, 2007, Schmitz- Hubsch et al, 2006a,b). The ICARS total score satisfied all psychometric criteria in a validation trial in patients with Friedrich's Ataxia (Cano et al., 2005; Metz et al., 2013) The ICARS has been used as an outcome measure in interventional studies in patients with ataxia, but there are no clinical data to suggest the magnitude of change on the ICARS that would correspond to a clinically relevant improvement, due to the absence of any drug producing such a benefit. The ICARS has been used in several studies in patients with Friedreich's Ataxia over the age of 8 years, in Phase 3 potentially pivotal trials (Lynch et al, 2010), as well as in open-label long-term studies (Meier et al, 2012).

The ICARS was also used in 2 interventional studies in children with A-T. Zanolli et al (2012) showed a statistically significant decrease in the ICARS total score in a placebo-controlled crossover trial in 13 children with A-T treated with oral betamethasone. Nissenkorn et al., 2013, using ICARS as a secondary endpoint, showed improvement in the static and kinetic subscales in a short-term open-label trial in 17 children (from 4 years of age) treated with amantadine.

Incoordination of eye movements, nystagmus, and loss in saccadic eye movement control is observed early in the A-T disease process (Hoche et al, 2012). The ICARS items relating to ocular motility do not provide a comprehensive assessment of this domain. Also, no scale has been validated for the measurement of ocular motility in A-T patients. The investigator will be asked to assess the ocular motility item based on the site's clinical paradigm and the instructions for rating the ICARS.

As A-T is a severe, relentlessly progressive disorder that leads to loss of independent movement in most patients by the age of 10 to16 years, any statistically significant difference from placebo over a 6-month period should be considered as a treatment success.

Although the ICARS will be administered in its entirety, results based on the Rescored mICARS (RmICARS) (refer to Table 4) will be the primary endpoint; RmICARS scores will be calculated based on the full ICARS assessment and used for the analysis of the primary endpoint.

The Modified ICARS (mICARS) excludes Items 8-12 related to kinetic function and Items 17-19 related to oculomotor functions that are required for visual control, as these items are not directly predictive of change in functioning. The Rescored mICARS (RmICARS) is re-scored by collapsing categories within specific items; this results in a smaller total sum score across the ICARS domains. The details of this rescoring will be available in the trial SAP.

In the IEDAT-04-2022 trial, the full ICARS scale will be administered by sites' qualified and trained local raters. Local ICARS raters will be neurologists identified by the PIs based on their experience and then qualified and trained to the administration of the ICARS in the context of the IEDAT-04-2022 by a specialized vendor. Details on the qualification and training process will be provided in a separate trial document.

Table 4: Comparison of ICARS, mICARS, and RmICARS (items and scores)

Full ICARS 100 points, 19 Items		**mICARS 54 points, 11 items**		**RmICARS 29 Points, 9 items**	
Posture and Gait Disturbance (34 points)		Posture and Gait Disturbance (34 points)		Posture and Gait Disturbance (23 points)	
1. Walking capacities	0-8	1. Walking capacities	0-8	1. Walking capacities	0-4
2. Gait Speed	0-4	2. Gait Speed	0-4	2. Gait Speed	0-3
3. Standing Capacities eyes open	0-6	3. Standing Capacities eyes open	0-6	3. Standing Capacities eyes open	0-4
4. Spread of feet eyes open	0-4	4. Spread of feet eyes open	0-4	4. Spread of feet eyes open	0-3
5. Body sway feet together eyes open	0-4	5. Body sway feet together eyes open	0-4	5. Body sway feet together eyes open	0-3
6. Body sway feet together eyes closed	0-4	6. Body sway feet together eyes closed	0-4	6. Body sway feet together eyes closed	0-3
7. Quality of sitting position	0-4	7. Quality of sitting position	0-4	7. Quality of sitting position	0-3
Kinetic Function (52 points) test left & right except drawing		Kinetic Function (12 points) test left & right except drawing		Kinetic Function (2 points) test left & right except drawing	
8. Knee tibia test (R/L)	0-4				
9. Action tremor (R/L)	0-4				
10. Finger to nose test (dysmetria) (R/L)	0-4				

Full ICARS 100 points, 19 Items		**mICARS 54 points, 11 items**		**RmICARS 29 Points, 9 items**	
11. Finger to nose test (intention tremor) (R/L)	0-4				
12. Finger to finger test (R/L)	0-4				
13. Pronation supination (R/L)	0-4	13. Pronation supination (R/L)	0-4		
14. Drawing	0-4	14. Drawing	0-4	14. Drawing	0-2
Speech Disorder (8 points)		Speech Disorder (8 points)		Speech Disorder (4 points)	
15. Fluency of speech	0-4	15. Fluency of speech	0-4		
16. Clarity of speech	0-4	16. Clarity of speech	0-4	16. Clarity of speech	0-4
Oculomotor Disorders (6 points)					
17. Gave evoked nystagmus	0-3				
18. Abnormalities of ocular pursuit	0-2				
19. Dysmetria of the saccade	0-1				
Total	0-100	Total	0-54	Total	0-29

ICARS = International Cooperative Ataxia Rating Scale; mICARS = modified International Cooperative Ataxia Rating Scale; RmICARS = Rescored modified International Cooperative Ataxia Rating Scale

7.7.2 *CGI-S and CGI-C*

The Clinical Global Impressions scale is a well-established research rating tool applicable to several disorders (Busner and Targum, 2007). The CGI was developed for use in National Institute of Mental Health-sponsored clinical trials to provide a brief, stand-alone assessment of the clinician's view of the patient's global functioning before and after initiating a trial medication (Guy, 1976). The CGI provides an overall clinician-determined summary measure that takes into account all available information, including a knowledge of the participant's history, psychosocial circumstances, symptoms, behaviour, and the impact of the symptoms on the participant's ability to function. The CGI actually comprises 2 companion one-item measures evaluating the following: (a) severity of illness from 1 to 7 and (b) change from the initiation of treatment on a similar 7-point scale. The first measure is the CGI-S, while the second one is the CGI-C or Improvement. In clinical research, the CGI is administered by an experienced clinician who is familiar with the disease under trial. Consequently, the CGI rater can make an expert clinical global judgment about the severity of the illness across various time points within the context of that clinical experience. The clinician makes a judgment about the total picture of the participant at each visit: the illness severity, the participant's level of distress and other aspects of impairment, and the impact of the illness on functioning. The CGI is rated without regard to the clinician's belief that any clinical changes are or are not due to medication and without consideration of the etiology of the symptoms (Busner and Targum, 2007).

To ensure independence of this rating from the rating of the ICARS (primary efficacy measure), a different rater (a physician, but not necessarily a neurologist) will perform the CGI assessments. The CGI rater will be initially identified by the PIs based on experience and then qualified and trained by a specialized vendor on the administration of the CGI. The CGI rater will be blinded to the treatment, safety assessments, and data, and will not have access to the ICARS scores in scoring the CGI. The CGI rater will not be allowed to administer the trial treatment.

Trial-specific instructions for the administration of the CGI-S/C, including a list of tools the raters must refer to while administering and scoring the scale, will be distributed to all the qualified CGI raters and must be followed throughout the trial to ensure consistency among raters and assessments.

7.7.2.1 CGI-S

The CGI-S scale measures global severity of illness on a 7-point scale and is the key secondary endpoint.

CGI-S will be performed at the Baseline Visit, Visit 6, and Visit 9. The change in CGI-S from baseline to Visit 9 will be classified as improved versus no change/worsened.

7.7.2.2 CGI-C

The CGI-C consists of a 7-point, clinician-rated, Likert-type scale assessing change from baseline. The CGI-C is evaluated on a 7-level ordinal scale, with categories ranging from "very much improved" as the best category (Category 1) to "very much worsened" as the worst category (Category 7). The mid-point of the scale is "no change" (Category 4). The CGI-C will be conducted at Visit 6 and Visit 9. The CGI-C will be assessed relative to the baseline assessment of the CGI-S. A video of the CGI-S interview and notes from the baseline assessment will be used to recall the baseline and anchor CGI-C to participants' initial status, allowing for a precise recall of participants' status by the rater. Given the comprehensive nature of the CGI-C assessment, all nuances of changes in participants' status will be captured.

7.7.3 *QoL Scale (EQ-5D-Y)*

Assessment of the QoL will be based on the EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older.

Improvements in QoL are an important goal for treatment of participants with A-T. To capture potential changes in the QoL in the NEAT trial, the assessment of the QoL will be based on the EQ-5D-Y, a preference-based measure of health status that is widely used around the world in clinical trials, population studies, and real-world clinical settings (EuroQol Research Foundation. EQ-5D-Y User Guide, 2020. Available from: https://euroqol.org/publications/user-guides); this tool is a validated version of the EQ-5D for younger respondents.

The EQ-5D is a standardized instrument for assessing health-related QoL in a variety of health conditions (Rabin and de Charro, 2001). The EQ-5D includes 5 levels of severity for each of the 5 EQ-5D dimensions (Herdman et al, 2011) and has been validated in a diverse patient population in 6 countries, including 8 patient groups with chronic conditions and a student cohort (Janssen et al, 2013). The EQ-5D-Y Interviewer Administered Proxy version 1 descriptive system comprises the same 5 dimensions as the standard EQ-5D, but uses more appropriate, child-friendly wording, and the visual analogue scale (VAS) (Section 12.3).

8 TRIAL OBJECTIVES

8.1 Primary Efficacy Objective:

To evaluate the effect of eDSP on CNS symptoms, as measured by the change in the RmICARS from baseline to Visit 9 compared to placebo in A-T (6- to 9-year-old participants primary analysis population). *[The RmICARS is a re-scored version of the ICARS and consists of 9 items across 3 domains (kinetic domain and speech limited to one item each), totaling a maximum score of 29 points.]*

8.2 Key Secondary Efficacy Objective:

To evaluate the overall clinical effect of eDSP, compared to placebo, in A-T (6- to 9-year-old participants primary analysis population), based on:

- CGI-S change from baseline to Visit 9.

8.3 Other Secondary Objective:

- CGI-C at Visit 9.

8.4 Exploratory Objectives:

To evaluate the effect of eDSP on QoL, compared to placebo, in A-T (6- to 9-year-old participants primary analysis population), based on:

- Health-related QoL using the change from baseline to Visit 9 of the domains in the EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older
- Health-related QoL using the change from baseline to Visit 9 of the EQ-5D-Y total score and VAS.

8.4.1 *Participants Aged 10 Years and Older:*

These primary/secondary/exploratory efficacy objectives will also be evaluated for participants aged 10 years and older. These analyses will be performed as data summaries in this older age cohort.

8.5 Safety Objective:

To evaluate the safety and tolerability of eDSP compared to placebo in participants with A-T, based on the occurrence of TEAEs, including SAEs and discontinuations due to AEs, and changes in vital signs, laboratory parameters, ECGs, and physical/neurological examination findings.

Safety analysis will be performed on the total population, on the 6- to 9-year-old participants, and on participants ≥10 years old.

9 INVESTIGATIONAL PLAN

9.1 Trial Design

This is an international, multi-center, randomized, double-blind, placebo-controlled, Phase 3 trial, designed to assess the effect of EryDex (DSP in autologous erythrocytes) administered by IV infusion once every 28 days (window +2 days, -7 days), on neurological symptoms of participants with A-T.

Listings of the trial procedures to be performed at each visit are provided in Section 11.

9.1.1 *End of Trial*

The end of the clinical trial (EOT) is defined as the date of the last visit of the last participant on Visit 9 in the trial.

9.2 Trial Population

9.2.1 *Inclusion Criteria*

To be eligible, the participant must meet all of the following criteria:

1. Participant meets clinical criteria for diagnosis of A-T. The neurological signs of A-T (incoordination of the head and eyes in lateral gaze deflection, gait ataxia associated with an inappropriately narrow base) must be documented. Such signs of A-T illustrate the body systems in which changes shall be confirmed, but the listed signs are examples, and other changes in those systems may be observed and documented to confirm the diagnosis of A-T.
2. Participant is in autonomous gait or is helped by periodic use of a support (i.e., ICARS score for *Item 1 – Walking Capacities* between 0 and 4 included).
3. Participant is at least 6 years of age (Dose 1 must be on or after date of 6th birthday), of either sex.
4. Genetic confirmation of A-T.
5. Body weight ≥15 kg.
6. The participant and parent/caregiver (if below the age of consent), or a legal representative, has provided written informed consent to participate. If consent is provided solely by the caregiver in accordance with local regulations, the participant must provide assent to participate in the trial, to the extent possible.

9.2.2 *Exclusion Criteria*

To be eligible, the participant must not meet any of the following criteria:

General

1. A disability that may prevent the participant from completing all trial requirements.
2. Current participation in another clinical trial. Participation in observational, non-interventional studies is allowed with approval by the Medical Monitor as long as trial investigational endpoint raters can remain blinded to the assessments from other studies, and as long as the other trial participation does not interfere with participation in this trial.

Medical History and Current Status

3. Immune impairment that includes CD4+ lymphocytes count <400/mm^3 (for participants less than 7 years old) or <150/mm^3 (for participants ≥7 years old). In presence of oral infections, like oral

candidiasis, documented at screening or recurrent as per medical history documentation, the limit increases to <200/mm^3 (for participants ≥7 years old).
4. History of severe impairment of the immunological system such that steroid treatment would be contraindicated.
5. Loss/removal of 250 mL or more of blood within the past 4 weeks prior to screening.
6. Current neoplastic disease or previous neoplastic disease not in remission for at least 2 years.
7. Severe or unstable pulmonary disease that impacts participant participation in the trial, in the opinion of the Investigators.
8. Uncontrolled diabetes. Participants with diabetes that has been stabilized (i.e., no hypoglycaemic or hyperglycaemic episodes in the past 3 months) will be eligible.
9. Any other severe, unstable, or serious disease or condition that in the Investigator's opinion would put the participant at risk for imminent life-threatening morbidity, need for hospitalization, or mortality.
10. Any clinically significant abnormality on standard laboratory examinations (haematology, biochemistry, urinalysis) at screening that remains abnormal on repeat testing, if considered as a possible sign of a clinical condition putting the participant at risk if enrolled. Eligibility of participants with abnormal laboratory test values will be determined by the Investigator in consultation with the Medical Monitor.
11. Participant with an early morning plasma cortisol level below 3-5 µg/dL (depending on assay), or participant exhibits signs or symptoms of adrenal insufficiency (Appendix 7), with an early morning plasma cortisol level below 10 µg/dL, and fails the ACTH stimulation test at screening.
12. Confirmed hemoglobinopathies (e.g., haemoglobin C disease, sickle cell anaemia, hereditary spherocytosis, or thalassemia).
13. Current chronic or acute significant renal and/or hepatic impairment that in Investigator's opinion will impact participant participation in the trial.
14. Participants with suicidal ideation.
15. Females who are pregnant or breast feeding. Females of childbearing potential using an adequate birth control method, as determined by their healthcare provider, will be eligible. For further details on the adequate contraceptive measures, please refer to Section 14.7.

Prior/Concomitant Medication

16. Any previous oral or parenteral steroid use within 6 weeks before Baseline. Treatment with inhaled or intranasal steroids for asthma or allergies, as well as use of topical steroids will be permitted.
17. Chronic condition or prior allergic reaction representing a contraindication to the use of dexamethasone or other steroid drugs.
18. Has participated in any other trial with an investigational drug and received a dose within 30 days or at least 5 half-lives (whichever is greater) prior to the Screening visit.
19. Has participated in a previous trial with EryDex treatment.
20. Requires any concomitant medication prohibited by the protocol, please refer to Section 11.5.

9.3 Documentation of Randomization

This is a randomized, double-blind, placebo-controlled trial in which a minimum of 86 participants aged 6 to 9 years old meeting all of the inclusion/exclusion criteria will be randomized equally (1:1; 43 participants per group) to one of 2 groups:

- **Group 1** – encapsulated DSP
- **Group 2** – encapsulated Placebo

To ensure the enrollment of the primary analysis population, the total number of participants 10 years of age and above is limited to 1 per site without prior approval from Sponsor (approximately 20 participants 10 years of age and above, 10 per treatment group, may be enrolled).

The randomization will be stratified by age (6 to 9 and ≥10 years), sex (male, female), and by geographic region.

Randomization will be done through a centralized Interactive Web Response System (IWRS). Each participant will receive 6 infusions of eDSP or placebo, given at monthly intervals. After 6 months of treatment, participants may receive eDSP treatment in an OLE trial.

At the time of screening, the participant will receive a unique 7-digit participant alphanumeric code consisting of 3 digits (2 letters and 1 number) representing the center and 4 digits numbers representing the order in which the participant was screened. For example, participant number AB1-0003 would correspond to the third participant screened at Center AB1. This participant alphanumeric code will be used for the participant throughout the remainder of their participation in the trial and will not be re-assigned to any other participant.

Only participants who meet all of the inclusion criteria and none of the exclusion criteria at Baseline will be eligible for being assigned to treatment. Assignment to treatment group will be done at Baseline, just prior to the time of dosing. At the time of randomization, the participant will be assigned a unique randomization number by the IWRS, which will be linked to the participant's treatment assignment. This number will be entered in the eCRF at Baseline but will not be used as a participant identifier.

9.4 Premature Discontinuation

Participants who discontinue from the trial treatment and the trial prematurely must have their reason for discontinuation entered in the Case Report Forms (CRFs) and in the source documents. Participants who discontinue from the trial after having received a dose of trial treatment will not be replaced.

All participants who discontinue prematurely prior to Visit 9 (final visit) will be asked to return for all final (Early Withdrawal/Visit 9) efficacy and safety evaluations.

For those participants who discontinue from the trial treatment prematurely, an attempt to perform the screening for adrenal insufficiency, via early morning (before 8:00 AM) plasma cortisol testing, should be made as soon as possible following discontinuation of the trial treatment, according to the procedures described in Appendix 7.

9.5 Record of Trial Participants and Screening Failures

The investigator will be required to maintain a confidential record of all trial participants, including all participants who were screened for the trial, but were not randomized to treatment. The confidential record must include sufficient information so that it would be possible for the investigators to contact the trial participant. Information on participants who have signed the Informed Consent Form (ICF)/Assent, but have failed screening, should be entered on the Screen Failure CRF.

The primary reason for screen failure will be recorded using the following categories:

- Did not meet entry criteria
- Major protocol deviation
- Pre-treatment Event/ AE
- Lost to follow-up

- Voluntary withdrawal (specify reason)
- Trial termination
- Other (specify reason)

Participant numbers assigned to participants who fail screening will not be reused.

10 TRIAL MEDICATION

10.1 Description, Labeling and Packaging

10.1.1 *Description of the Supplies*

The DSP sterile solution will be used for Group 1. [redacted] in water for injection will be used as placebo, as it contains the non-active principal sodium chloride, and its osmolality is identical with that of the DSP solution that is used in the EDS process for Groups 1. All participants deemed suitable for inclusion in the trial will be treated with either EryDex or the autologous RBCs processed using placebo for injection instead of DSP solution (Group 2).

Table 5: Investigational Medicinal Product Composition

[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]

The active ingredient used for the ampules is dexamethasone sodium phosphate.

Table 6: [redacted]

[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]

10.1.2 *Stability of the Product*

Stability studies have demonstrated that the drug product is stable for at least 3 years if stored at 2°C to 8°C and not exposed to light.

10.1.3 *Packaging*

The primary packaging for Group 1 (Active with investigational medicinal product [IMP]) consists of a [redacted] The secondary packaging is a cardboard box with one ampule inside. One ampule will be needed for preparing each dose of EryDex. For Group 1, [redacted] For Group 2, (placebo), the primary and secondary packaging will be the same as for the IMP, but containing [redacted] Trial medication will be labelled and assigned to participants such that trial site pharmacy staff will be blinded to treatment group as well. No opened ampule should be reused, and damaged ampules must not be used.

10.1.4 *Labeling*

10.1.4.1 Dexamethasone Sodium Phosphate

Labeling will be done in a manner consistent with the trial design, according to the local requirements. Each box will be labelled with a twin label with a tear-off portion. The tear-off part of the twin label must be attached to the Prescription Form.

The outer packaging will contain the following information in the local language: Investigational New Drug (IND) number, EU Clinical Trial (EUCT) number, Integrated Research Application System (IRAS) ID, trial number, site number, participant number, Kit ID number, quantity, content, dosage form and route of administration, Sponsor's name and contact information (address and telephone number), storage conditions (2°C to 8°C), lot number, expiry date, and warnings.

The single ampules will be labelled with the following information in the local language: trial number, Kit ID number, lot number, expiry date, participant number, quantity, content, dosage form and route of administration, warning, and Sponsor name.

10.1.4.2 Placebo

Labeling will be done in a manner consistent with the trial design, according to the local requirements. Each box will be labelled with a twin label with a tear-off portion. The tear-off part of the twin label must be attached to the Prescription Form.

The outer packaging will contain the following information in the local language: IND number, EUCT number, IRAS ID, trial number, site number, participant number, Kit ID number, quantity, content, dosage form, and route of administration, Sponsor's name and contact information (address and telephone number), storage conditions (2°C to 8°C), lot number expiry date, and warnings.

The single ampules will be labelled with the following information in the local language: trial number, Kit ID number, lot number, expiry date, participant number, quantity, content, dosage form and route of administration, warning, and Sponsor's name.

10.2 Dose Encapsulation Process and Administration

Participants will be randomized to receive treatment with eDSP (Group 1) or Placebo (Group 2)

- Group 1 will receive an EryDex IV infusion of [redacted] (mean ± SD) of eDSP; this is the average dose from the analysis of the samples of high-dose group participants in the ATTeST trial who received the same loading dose of DSP to be used in IEDAT-04-2022). The infusion will be a total volume of approximately [redacted] (final volume of the bag minus the volume transferred into the satellite sample bag) and will be given over approximately [redacted].
- Group 2 will receive an EryDex IV infusion of encapsulated Placebo in a total volume of approximately [redacted] (final volume of the bag minus the volume transferred into the satellite sample bag) given over approximately [redacted].

The above IV infusions with either encapsulated DSP or Placebo previously taken from the same participant will be prepared for each group, using the EDS process 3.3.2 (or an equivalent version with the same performance and safety). [redacted]

[redacted]

The caregiver/participant should be advised to comply with the visit schedule. If the participant is unable to have the infusion performed at the scheduled time, it should be done as soon as is feasible. No infusion should be performed fewer than 14 days after the prior infusion.

Possible AEs that might occur related to the infusion include vaso-vagal reactions, rash, fever or anemia. The likelihood of transfusion reactions is very low, since autologous RBCs are being infused.

[redacted]

10.3 Storage

Dexamethasone sodium phosphate is provided in ampules [redacted] to be stored at 2°C to 8°C and protected from light. Storage temperatures will be recorded by a data-logger and kept on trial file. In case of temperature excursions beyond the limits set by the manufacturer, immediate information should be given to the Sponsor, or to the Clinical Research Associate for the appropriate actions to be taken.

Ampules of DSP or placebo assigned to a participant must not be used again for other participants; after use, these may be stored at room temperature until accountability is assessed. Upon approximation of the expiry date, and after accountability is performed by the Clinical Research Associate, the used/unused ampules of DSP or placebo will be destroyed at the site following local procedures for the disposition of drugs.

10.4 Blinding and Randomization

This is a randomized, double-blind trial; therefore, the Sponsor/CRO personnel (other than the one assigned to unblinded role and responsibilities), Investigator, site staff, and participant will not be aware of the treatment assignments.

A minimum of 86 participants 6 to 9 years old will be randomly assigned equally to 1 of the 2 treatment groups (1:1; 43 participants/ group) by the Investigator using an IWRS. To ensure the enrollment of the primary analysis population, the total number of 10 years of age and above is limited to 1 per site without prior approval from Sponsor (approximately 20 participants 10 years of age and above, 10 per treatment group, may be enrolled). The randomization will be stratified by age (6 to 9 and ≥10 years), sex (male, female), and by geographic region. Participants will be considered to have completed the trial when the final endpoint assessment at Visit 9 has been performed.

The customized IWRS will be able to capture, store, and process data for all participants in this project. All data inputs by the user will be checked for validity by the IWRS whenever such entries are performed. The user will be notified of any invalid entries or transaction errors that could prevent the user from continuing with the transaction. The IWRS will store data in a validated database. The IWRS can be customized to provide additional messages at specific visits. If the users encounter system problems or lock the IWRS access, they will be presented with the option to contact a customer service representative at the IWRS Help Desk. The stratification algorithm will be customized for the protocol and fully tested within the IWRS.

The PI (or delegated back-up) may break the blind on an individual participant by using the IWRS. This should be done only in a medical emergency in which the participant's treatment assignment needs to be known in order to properly treat the participant. The reason for breaking the blind must be provided.

10.5 Accountability

During the course of the trial, the trial pharmacist or site personnel designated by the Investigator to manage the trial medication must record the trial treatment disposition and keep the accountability forms updated. The trial treatment accountability forms will be cross-checked with the EryDex checklist. A copy of the accountability forms must be kept in the trial files at the site, and the other copy will be withdrawn by the CRO staff responsible for monitoring the trial medication. The used and unused dexamethasone and placebo ampules, as well as the EryDex checklist, will be kept at the site for accounting and reconciliation by the Clinical Research Associate versus the trial documentation. The used infusion bags containing any remaining erythrocytes (with EryDex) will be disposed of at the site, with appropriate documentation.

10.6 Overdose

Participants in the active group in this trial will receive up to 6 infusions of EryDex. Each infusion should contain a maximum of [redacted] DSP encapsulated in autologous RBCs. However, based upon results of prior studies, some inter-individual variability in loaded dose is expected; therefore, administered doses of DSP greater than [redacted] will be considered an overdose. Treatment for an overdose should be the same as the clinical management for an overdose of dexamethasone.

10.7 Occupational Safety

There are no risks anticipated related to the trial medication for the staff involved in administering EryDex to the participants in this trial. Standard procedures and precautions for handling needles and biological samples should be followed.

11 EVALUATIONS AND PROCEDURES

11.1 Written Informed Consent

a) Prior to the initiation of any screening procedure, every participant and parent/caregiver (if below the age of consent), or a legal representative must provide written consent and sign the ICF; in accordance with local regulations, children must provide assent to participate in the trial, to the extent possible according to the procedure described in Section 17.1. The details of the trial should be discussed with the participant and parent/caregiver or legal representative (if necessary) prior to obtaining informed consent, and the ICF and Assent Form must be signed and dated by the participant, parent/caregiver/ legal representative (if necessary), and by the Investigator or their designee. A copy of the signed ICF will be provided to the participant, and the original will be retained with the source documents.

b) A separate section of the ICF will be used to obtain specific consent for the optional sample for biomarker development.

c) Participants who complete the current trial and elect to receive open-label extension treatment with eDSP will need to sign a separate consent form.

11.2 Trial Conduct

This trial is divided into 3 periods: a Screening period, an approximately 6-month Treatment Period (Visits 1-8), and a Trial Completion/Early Withdrawal Visit (Visit 9).

11.2.1 *Screening Period*

After providing consent/assent, as described in Section 11.1, each participant will undergo a 30-day Screening Period to evaluate their eligibility for the trial.

During the screening period, any previous treatments with other corticosteroid compounds will be withdrawn (washout from previous treatment); such washout from previous treatment may (depending on the dose of the previous corticosteroid) result in a prolongation of the standard 30-day screening period to 45 days from the completion of the tapering off, which, in this specific case will not constitute a deviation from the Protocol. Following informed consent, Investigators should consult with the Medical Monitor about such specific cases.

If any abnormal laboratory test results, vital sign measurements, or ECG findings of clinical significance are noted at screening, these must be repeated during the 30-day screening period and the results made available prior to making the final decision on a participant's eligibility at Baseline. Adverse events, reported by the participant or observed by the investigator, and the use of concomitant medication will be recorded from the time of signing of informed consent through the end of the trial.

The initial rating of the ICARS will be performed at the screening visit. Throughout the trial, the ICARS rating will be performed by qualified raters trained in the use of the scale and not involved in rating other scales. The ICARS rater will not have access to the results of other scales or to the safety data. The ICARS rater cannot administer the trial treatment. ICARS ratings must be completed without consulting scores from the previous visit.

11.2.2 *Treatment Period*

At Baseline (Visit 1), additional pre-treatment evaluations will be performed as well as collection of baseline safety information. AEs and concomitant medications will be assessed. The inclusion/exclusion criteria will be fully reviewed and participants meeting all criteria will be randomized to 1 of the 2 treatment groups.

The baseline efficacy ratings of the ICARS, CGI-S (videotaped), and EQ-5D-Y will be performed prior to dosing. At each applicable visit, the ICARS should be the first scale administered, followed by the CGI-S/CGI-C, and then by the EQ-5D-Y (please refer to Table 1 for the list of tests to be conducted at each visit). Neurological assessments must be performed before any phlebotomy or IV insertion so that upper extremity neurological exam is not impeded by an IV line, except at screening and Visit 9.

[redacted]

Visits 2 and 3 will be conducted by phone (unless there are safety concerns) to assess AEs and concomitant medications.

Participants will come to the investigational sites at Baseline and at Visits 4, 5, 6, 7, and 8 to receive trial treatment and to undergo trial safety assessments. Each treatment must be scheduled every 28 days (window +2 days, -7 days), calculated from the previous infusion. The window between an infusion and the subsequent one should be kept as regular as possible throughout the trial, avoiding fluctuations in administration windows. In any case, infusions should not be given fewer than 14 days apart. Any instances where it is not possible to administer the infusion within the designated window should be documented as a protocol deviation and the Investigators should immediately contact the Medical Monitor to discuss if the participant may continue in the trial, and if so, to agree on the path forward that will best ensure the participant's safety and meet the trial's goals.

Due to the length of time required for the encapsulation procedures (>2 hours), Investigators should define the number of participants that can be visited and dosed on the same day, based on the site staff availability and RCL status.

At Visit 6, in addition to safety assessments, efficacy assessments (ICARS, EQ-5D-Y, CGI-S, CGI-C) will be performed before treatment administration.

11.2.3 *Trial Completion/Early Withdrawal Visit*

The final evaluations will take place at Visit 9 (or early withdrawal, if a participant discontinues the trial treatment or trial prematurely) and will include all efficacy (ICARS, CGI-S, CGI-C, and EQ-5D-Y) and safety assessments. The efficacy evaluations at Visit 9 will be used as endpoint assessments for statistical purposes.

At this visit, the occurrence of any AEs or SAEs reported by the participant/caregiver or observed by the Investigators since the previous visit will be recorded.

Throughout the trial, Investigators may perform any additional safety assessments deemed necessary to assess subjects' safety. The medical management of a patient, outside of the requirements of the protocol, is within the discretion of the Principal Investigator and Principal Investigator's delegated medical team.

Participants who discontinue prematurely will still be asked to perform this visit.

Participants who complete the trial's full treatment period, and complete the trial assessments may elect to receive eDSP treatment in an OLE trial.

An overview of the schedule of assessments for the trial is presented in Table 1.

11.2.4 *Screening Period*

At Screening each participant will report to the clinic where the following procedures will be performed to establish eligibility for the trial:

a) Obtaining written informed consent (before any trial procedures) by participants and parent/caregiver (if below the age of consent), or a legal representative. In accordance with local regulations, children must provide assent to participate in the trial, to the extent possible
b) Demography
c) Medical history
d) Physical and neurological examination
e) Vital signs – body weight and height, calculation of body mass index (BMI), temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic blood pressure (BP), and respiratory rate
f) C-SSRS
g) Laboratory evaluations comprising the following tests:
   - hematology - RBC, white blood cells (WBC), hemoglobin, hematocrit, platelets, neutrophils, eosinophils, basophils, lymphocytes, monocytes, mean corpuscular volume , mean corpuscular hemoglobin , mean corpuscular hemoglobin concentration , and RBC distribution width
   - clinical chemistry – total protein, albumin, bilirubin, aspartate aminotransferase, alanine aminotransferase, blood urea nitrogen, serum creatinine, sodium, potassium, chloride, bicarbonate, calcium, phosphorus, serum iron, lactate dehydrogenase (LDH), alkaline phosphatase, glucose, creatine phosphokinase, triglycerides, and cholesterol (total, high-density lipoprotein, and low-density lipoprotein)
   - urinalysis (automated) – color, pH, specific gravity, glucose, ketones, nitrites, protein, bilirubin, hemoglobin, urobilinogen, and reflex microscopic RBC, WBC, and casts (if indicated)
h) Special laboratory tests: CD4+ lymphocytes count (a local laboratory test), α–fetoprotein, CRP
i) Females will have a serum pregnancy test
j) 12-lead standard ECG
k) Prior (previous 6 weeks) and concomitant medications
l) AEs occurring after giving informed consent, during the screening period, and before the first dose of trial treatment
m) Plasma cortisol – sample to be collected before 8:00 AM during the screening period and prior to randomization. If the 8:00 AM cortisol level is within the reference normal range, the participant can be enrolled in the trial. If the 8:00 AM cortisol level is below 3-5 µg/dL (depending on assay) regardless of symptoms, or the participant exhibits signs or symptoms of adrenal insufficiency (see Appendix 7) and has a cortisol <10 µg/dL, the participant will receive a high-dose ACTH stimulation test as soon as possible. If the ACTH stimulation test is normal, the participant can be enrolled after the effects of the ACTH have dissipated (e.g., 30 days after ACTH administered, resulting in a prolongation of the Screening Period). If the participant fails the ACTH stimulation test, they will be excluded from the trial and referred to an endocrinologist (a pediatric endocrinologist, depending on the participant's age) with a recommendation to prescribe stress dose steroids.
n) ICARS

o) Participant eligibility and inclusion/exclusion criteria check (**all test results documenting the eligibility, even in case of repeated tests because of abnormalities at screening, must be available on or before Visit 1**).

If any abnormal laboratory test results, vital sign measurements, or ECG findings of clinical significance are noted at the screening visit, these must be repeated during the 30-day Screening period and the results made available prior to making the final decision on a participant's eligibility for the trial at Baseline. AEs, reported by the participant or observed by the Investigators, and the use of concomitant medication will be recorded from the time of signing of informed consent through the end of the trial.

Participants who initially fail eligibility criteria for the trial may be re-screened in cases where the PI, in consultation with the Medical Monitor, believes this is appropriate based upon an understanding of the health condition of the participant.

11.2.5 *Visit 1: Baseline and Dose 1*

The Baseline visit is designed as a single-day visit but, in case of logistical issues for a particular participant, the visit can be performed over 2 days.

Baseline assessments to be performed (pre-dose) are as follows:

a) Inclusion and exclusion criteria check: review of all selection criteria
b) Vital signs (within 15 minutes before the IV infusion of the trial treatment) - body weight and height, calculation of BMI, temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate
c) Neurological examination
d) 12-lead standard ECG *
e) Assessment of physical development, sexual maturation, and the effect of trial treatment on these aspects (Tanner scale)
f) Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to infusion
g) C-SSRS
h) Bone mineral density, where country regulations allow
i) ICARS
j) CGI-S (videotaped)
k) EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older
l) Review of AEs – reported by the participant or observed by the Investigator, that occurred before the first dose of trial treatment
m) Review of concomitant medication
n) Routine laboratory evaluations* on the diverted blood sample (see Section 11.3), comprising the following tests:
   - hematology
   - clinical chemistry
   - urinalysis
o) Special laboratory tests on the diverted blood sample (see Section 11.3): CD4+ lymphocytes count (a local laboratory test), CRP
p) An optional blood sample for biomarker development will be obtained for participants who consent to this procedure on the diverted blood sample (see Section 11.3)
q) Following randomization using IWRS, the trial treatment will be prepared and administered immediately after the completion of the EryDex process, as follows:

- [redacted] blood collected after blood diversion (see Section 11.3) for aerobic culture
- [redacted] blood collected after blood diversion and sampling for culture, for use in the EryDex process
- Addition of randomized trial treatment (DSP or placebo) to the EDS
- [redacted]
- Trial treatment administration by IV infusion, TO BE STARTED within 30 minutes after the EDS process has been completed
- The satellite sample bag will be detached and used for sterility culture tests: a sample of EryDex (approximately 1 mL per inoculum for a total of 2 mL) will be collected to perform a sterility test using a culture-based method as presented in Appendix 5. A 1-mL sterile sample of EryDex will be stored under refrigeration as a "Retention Sample". Appendix 3.

r) Once the final bag is empty (EryDex has been infused), flush the infusion lines with normal saline only as per clinical practice (e.g., by infusion pump or by gravity)Post-infusion assessments:
- Vital signs, within 15 minutes after the infusion - temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate
- Review of AEs – reported by the subject or observed by the Investigator
- Review of concomitant medication

Physical examination must be conducted on the day of study drug administration.

*These routine laboratory and ECG evaluations will be repeated only if abnormalities requiring follow- up were noted on the Screening evaluations; results from these repeat assessments must be available at baseline to confirm eligibility before the subject can be randomized to treatment. If abnormalities are noted on the screening ECG, then the ECG performed as part of Visit 1 should be done in triplicate.

11.2.6 *Visit 2 (24 hours after Visit 1) - by phone or in person*

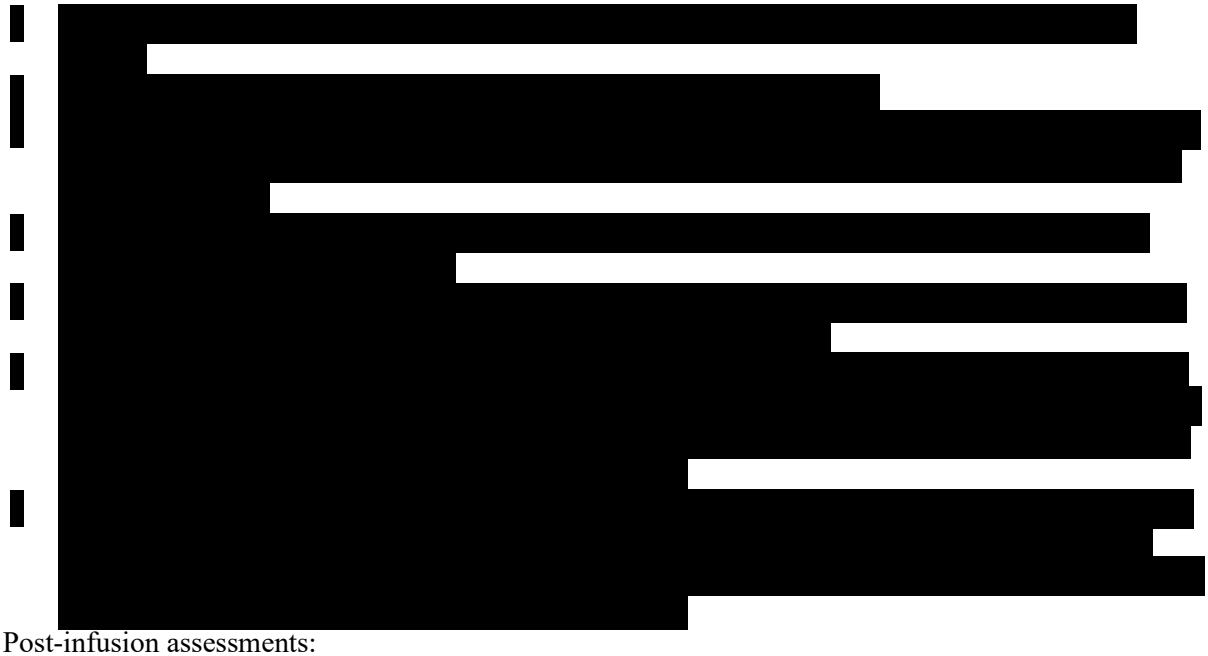
a) Review of AEs – reported by the subject or observed by the Investigator
b) Review of concomitant medication

11.2.7 *Visit 3 (Day 14) - by phone or in person*

a) Review of AEs – reported by the subject or observed by the Investigator
b) Review of concomitant medication

11.2.8 *Visit 4 Dose 2*

a) Vital signs (within 15 minutes before the IV infusion of the trial treatment) – including body weight and height
b) Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to infusion.
c) C-SSRS
d) Review of AEs – reported by the participant or observed by the Investigator
e) Review of concomitant medication
f) Study treatment will be prepared and administered, immediately after the completion of the EryDex process, as follows:
[redacted]



g) Post-infusion assessments:
   - Vital signs, within 15 minutes after the infusion: temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate
   - Review of AEs – reported by the participant or observed by the Investigator
   - Review of concomitant medication

   Physical examination must be conducted on the day of trial treatment administration.

   *These routine laboratory and ECG evaluations will be repeated only if abnormalities requiring follow-up are noted on the Screening evaluations; results from these repeat assessments must be available at Baseline to confirm eligibility before the participant can be randomized to treatment.

11.2.9 *Visit 5 Dose 3*

a) Vital signs (within 15 minutes before starting the IV infusion of the trial treatment) – including body weight and height
b) Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to infusion
c) C-SSRS
d) Review of AEs – reported by the participant or observed by the Investigator
e) Review of concomitant medication
f) Trial treatment will be prepared and administered, immediately after the completion of the EryDex process, as follows:

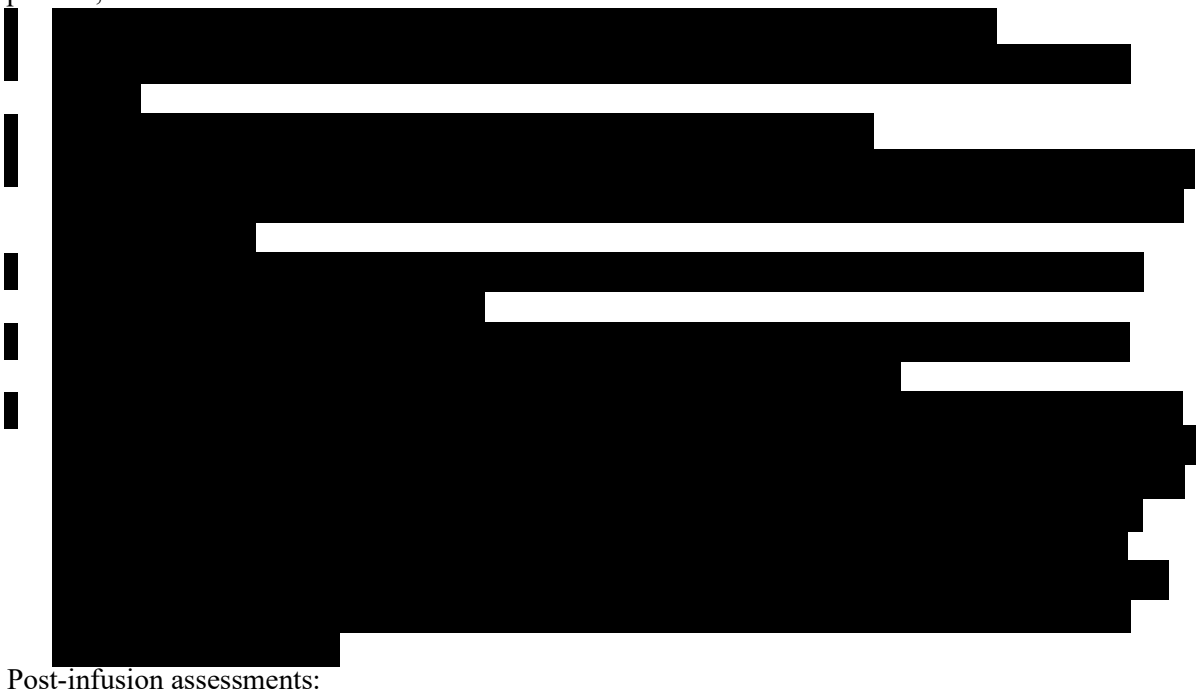
g) Post-infusion assessments:
   - Vital signs, within 15 minutes after the completion of the infusion – temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate
   - Review of AEs – reported by the subject or observed by the Investigator
   - Review of concomitant medication

   Physical examination must be conducted on the day of study drug administration.

11.2.10 *Visit 6 Dose 4*

a) Neurological examination
b) Vital signs (within 15 minutes before starting the IV infusion of the trial treatment) – including body weight and height
c) Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to infusion.
d) C-SSRS
e) ICARS
f) CGI-S
g) CGI-C
h) EQ-5D-Y Interviewer Administered Proxy version 1 for subjects 6 to 9 years of age and self-reported for subjects 10 years of age and older
i) Review of AEs – reported by the participant or observed by the Investigator
j) Review of concomitant medication
k) Routine laboratory evaluations on the diverted blood sample (see Section 11.3), comprising the following tests:
   - haematology
   - clinical chemistry
   - urinalysis
l) Special laboratory tests, on the diverted blood sample (see Section 11.3): CD4+ lymphocytes count (a local laboratory test), CRP
m) An optional blood sample for biomarker development will be obtained for subjects who consent to this procedure, on the diverted blood sample (see Section 11.3)

n) Trial treatment will be prepared and administered, immediately after the completion of the EryDex process, as follows:



o) Post-infusion assessments:
- Vital signs, within 15 minutes after the completion of the infusion – temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate
- Review of AEs – reported by the participant or observed by the Investigator
- Review of concomitant medication

Physical examination must be conducted on the day of trial treatment administration.

11.2.11 *Visit 7 Dose 5*

a) Vital signs (within 15 minutes before starting the IV infusion of the trial treatment) – including body weight and height
b) Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to infusion.
c) C-SSRS
d) Review of AEs – reported by the participant or observed by the Investigator
e) Review of concomitant medication
f) Trial treatment will be prepared and administered immediately after the completion of the EryDex process, as follows:

[redacted]

g) Post-infusion assessments:
- Vital signs, within 15 minutes after the completion of the infusion – temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate
- Review of AEs – reported by the subject or observed by the Investigator
- Review of concomitant medication

Physical examinations must be conducted on the day of trial treatment administration.

11.2.12 *Visit 8 Dose 6*

a) Vital signs (within 15 minutes before starting the IV infusion of the trial treatment) – including body weight and height
b) Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated prior to infusion.
c) C-SSRS
d) Review of AEs – reported by the subject or observed by the Investigator
e) Review of concomitant medication
f) Study treatment will be prepared and administered, immediately after the completion of the EryDex process, as follows:

[redacted]

[redacted]

g) Post-infusion assessments:
- Vital signs within 15 minutes after the completion of the infusion – temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate
- Review of AEs – reported by the participant or observed by the Investigator
- Review of concomitant medication

Physical examination must be conducted on the day of trial treatment administration.

11.2.13 *Visit 9/Early Withdrawal/Trial Completion (No Treatment)*

The following procedures will be performed at the final Visit 9, or if the participant discontinues from the trial treatment and trial prematurely:

a) Physical and neurological examination
b) Vital signs – body weight and height, temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate
c) Assessment of physical development, sexual maturation, and the effect of the administered treatment on these aspects (Tanner scale). Tanner staging of the breasts in pre-menarchal females and of the scrotum in males who have not completed puberty will be performed
d) Females noted to be at Tanner Stage 2 or greater at this Visit will have a serum pregnancy test
e) 12-lead standard ECG
f) ICARS
g) CGI-S
h) CGI-C
i) EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older
j) C-SSRS
k) Routine laboratory evaluations:
   - hematology
   - clinical chemistry
   - urinalysis
l) Special laboratory tests: CD4+ lymphocytes count (a local laboratory test), alpha–fetoprotein, CRP
m) Pregnancy test
n) Plasma cortisol – sample to be collected before 8:00 AM. If the 8:00 AM cortisol level is below 3-5 µg/dL (depending on assay) regardless of symptoms, or the participant exhibits signs or symptoms of adrenal insufficiency (Appendix 7) and has a cortisol <10 µg/dL, the participant will receive a high-dose ACTH stimulation test as soon as possible. Participants failing the ACTH stimulation test will be referred to an endocrinologist (a pediatric endocrinologist, depending on the participant's age) with a recommendation to prescribe stress dose steroids.
o) An optional blood sample for biomarker development will be obtained for participants who consent to this procedure
p) Bone mineral density, where country regulations allow
q) Review of AEs – reported by the participant or observed by the Investigator
r) Review of concomitant medication

11.2.14 *Unscheduled/As-needed Assessments*

Participants in whom there is a clinical suspicion of hemolysis (including pallor, lethargy, weakness, headaches, cold hands and feet, tachycardia, irregular heartbeats, heart murmur, shortness of breath, dizziness or lightheadedness, change in urine color, scleral icterus, jaundice or splenomegaly) during the trial may have the following testing performed depending on the clinical circumstances and as determined by the Investigators:

- Hemolysis Panel: free plasma hemoglobin, haptoglobin, bilirubin (total and conjugated), CBC, LDH and urinalysis
- RBC antibodies (immunoglobulin [Ig]G, IgM, Qualitative Direct Coombs test)
- Free plasma hemoglobin (1 hour after infusion)

In addition, Investigators can decide to perform any additional safety assessments deemed necessary to assess participants' safety throughout the trial. The medical management of a participant, outside of the requirements of the protocol, is within the discretion of the Principal Investigator and Principal Investigator's delegated medical team.

11.2.15 *Visit Windows*

Each treatment must be scheduled every 28 days (window +2 days, -7 days), calculated from the previous infusion. The window between an infusion and the subsequent one should be kept as regular as possible throughout the trial, avoiding fluctuations in administration windows.

Infusions should not be given in any case fewer than 14 days apart. Any instances where it is not possible to administer the infusion within the designated window should be documented as a protocol deviation and the Investigators should immediately contact the Medical Monitor to discuss if the participant may continue in the trial, and if so, to agree on the path forward that will best ensure the participant's safety and meet the protocol goals.

Table 1 provides the trial day windows in detail for each visit.

11.3 Laboratory Sample Collection

11.3.1 *Instructions for Aseptic Procedure*

Aseptic procedures have been implemented to ensure sterility during blood collection for the EryDex process, the various steps of the EryDex process, sample collection for laboratory tests and for sterility testing of EryDex, and administration of EryDex to the participant. Details of these procedures are described in a separate document entitled "Aseptic Procedure Guideline," which will be provided to each site. These procedures include the following measures that must be performed to ensure sterility:

- Blood sampling under sterile conditions.
- Careful hand washing and use of fresh (non-sterile) gloves for each participant, disinfected frequently using a bactericidal rub.

[redacted]

11.3.2 *Blood sampling*

[redacted]

11.3.3 *Urine collection*

Urine needed for the urinalysis will be collected at Screening, Visit 1 (optional), Visit 6 and Visit 9.

For females noted to be at Tanner stage 2 at Baseline, urine pregnancy tests will be performed monthly before every infusion.

All blood and urine samples for safety (hematology/ clinical chemistry/ urinalysis) and special laboratory evaluations will be analysed by an external (central) laboratory, with the exception of plasma cortisol, ACTH, and sterility tests. In certain circumstances the investigator may choose to utilize a local laboratory if there is an immediate need for the test results.

11.4 Measurement of Dexamethasone Sodium Phosphate and Other Analytes in Infusion Bag Sample

Once the EryDex process has been completed, a sample of the contents of the infusion bag will be collected for analysis of the DSP concentration, so that the actual dose administered to each participant can be determined. The procedure to be followed for this sample is as follows:

11.5 Concomitant Medications

All participants to be included in the trial must **not** have received **oral or parenteral steroid** therapy within 6 weeks prior to the administration of EryDex. However, treatment with inhaled or intranasal steroids for asthma or allergies, as well as the use of topical steroids, will be permitted. In addition, participants with a history of use or currently using any of the following medications will be excluded from participation, according to the guidelines specified:

- Narcotic analgesics – use within 6 weeks prior to baseline;
- Antipsychotics – unless used at a low, stable dose starting at least 6 weeks prior to baseline, in which case they will be permitted;
- Drugs that are strong inducers (e.g., carbamazepine, St. John's wort) or inhibitors (e.g., clarithromycin, grapefruit juice) of cytochrome P450 3A4 (CYP3A4) within 6 weeks prior to baseline;

Drugs that are inducers or inhibitors of CYP3A4 may alter the plasma levels of dexamethasone, which is metabolized by this enzyme. Therefore, in the current trial, any drug that is a strong inducer or inhibitor of CYP3A4 will be prohibited from use during the trial. Participants who are taking these drugs upon entry into the screening period should have the medication discontinued, if possible, or be switched to another similar medication that does not have this property. A list of drugs that are

strong inducers or inhibitors of CYP3A4 can be provided to the Investigator upon request and the Investigator should refer as well to the local prescriber approved label for dexamethasone.

- Amphotericin-B within 6 weeks prior to baseline – combination with corticosteroids or corticotropin (ACTH) may induce hypopotassemia;
- Any immunization, vaccination or skin test, especially using a live, attenuated vaccine, within 4 weeks prior to screening. The use of vaccines that do not contain a live, attenuated virus (e.g. diphtheria, pertussis, and tetanus [DPT] vaccine, and coronavirus disease 2019 RNA vaccines) during the course of the trial is left to the clinical judgement of the PI, and the standard of care at the site.

Participants on stable doses of other drugs, such as antihypertensives, benzodiazepines, antihistamines (histamine receptor blockers), birth control, proton-pump inhibitors, vitamins/multi-vitamins, anti- diabetic agents, and lipid-lowering agents (e.g., statins), will be eligible for the trial. The use of other concomitant medications with CNS effects should be discussed with and approved by the Medical Monitor before prescribing to the participant during the trial. Use of immunoglobulins either administered by IV or intramuscular (IM) route is permitted; the Investigator should determine the optimal timing of the dose with respect to the EryDex infusion.

During the entire period of the trial, starting with the signing of the ICF, any new treatment that is initiated must be reported in the eCRF using the pharmacological name (not the trade name, with the exception of medicines in fixed combination), specifying daily dose, route, duration of treatment, reason for use – following the instructions included in the eCRF guidelines. After screening, participants should be instructed to contact the Investigator before starting any over-the-counter or prescription medication on their own or as prescribed by their physician. The Medical Monitor should be informed of any new medication that may be a prohibited medication and is started during the ,trial they will decide whether or not it is acceptable for the participant to continue in the trial.

12 EFFICACY TOOL AND EVALUATIONS

12.1 International Cooperative Ataxia Rating Scale

12.1.1 *Description of the ICARS*

The primary efficacy endpoint in this trial will be the mean change of the RmICARS from baseline to Visit 9 compared to placebo in 6- to 9-year-old participants with A-T.

The ICARS (Trouillas et al, 1997), the most frequently used clinician-(neurologist) rated measure in patients with ataxias, was developed by a Committee of the World Federation of Neurology to help standardize common manifestations of syndromes that lead to cerebellar dysfunction. The ICARS is a 100-point, semi-quantitative scale offering a compartmentalized quantification of the following 4 sub- scores: Posture and Gait Disturbances (34 points), Kinetic Functions (52 points), Speech Disorders (8 points), and Oculomotor Disorders (6 points).

The complete ICARS will be rated in the current trial; however, for the primary efficacy endpoint, the rescored 'Modified' ICARS will be used. The 'Modified' ICARS excludes all the Oculomotor Disorders items (items 17-19) and items 8-12 in the Kinetic Functions domain, as these items do not directly contribute to functioning. The Rescored mICARS (RmICARS) is re-scored by collapsing categories within specific items [redacted]; this results in a smaller total sum score across the ICARS domains. The details of this rescoring will be available in the trial SAP.

12.1.2 *Validity of International Cooperative Ataxia Rating Scale in Ataxia-telangiectasia Patient Population*

The internal consistency, criterion-related validity, and internal construct validity of the ICARS have been established in patients with focal cerebellar lesions (Schoch B et al, 2007). The inter-rater reliability, test-retest reliability and internal consistency have also been validated in patients with spinocerebellar ataxia (Weyer et al, 2007, Schmitz-Hubsch et al, 2006a,b). The ICARS total score satisfied all psychometric criteria in a validation trial in patients with Friedrich's Ataxia (Cano et al, 2005; Metz et al 2013) The ICARS has been used as an outcome measure in interventional studies in patients with ataxia, but there are no clinical data to suggest the magnitude of change on the ICARS that would correspond to a clinically relevant improvement, due to the absence of any drug producing such a benefit. The ICARS has been used in several studies in patients with Friedreich's Ataxia over the age of 8 years, in Phase 3 potentially pivotal trials (Lynch et al, 2010), as well as in open-label long-term studies (Meier et al, 2012).

The ICARS was used also in 2 interventional studies in children with A-T. Zanolli et al, (2012) showed a statistically significant decrease in the ICARS total score in a placebo-controlled crossover trial in 13 children with A-T treated with oral betamethasone. Nissenkorn et al., using ICARS as a secondary endpoint, showed improvement in the static and kinetic subscales in a short-term open- label trial in 17 children (from 4 years of age) treated with amantadine (Nissenkorn et al, 2013). As A-T is a severe, relentlessly progressive disorder that leads to loss of independent movement in most patients by the age of 10 to 16 years, any statistically significant difference from placebo over a 6-month period should be considered as a treatment success.

Most of the developmental and validation studies of the ICARS were performed in patients who were 10 years of age or older; an international trial has been completed (ClinicalTrials.gov: NCT01942850) to extend its validity to patients in the age-range of 5 to10 years. Data collected from the validation trial in selected centers [Data on file, EryDel S.p.A] indicated that the distribution of scores for the ICARS for patients with A-T under 10 years of age was similar to the scores for these patients on other validated scales

such as the Scale for Assessment of Rating of Ataxia (SARA) (Schmitz-Hubsch et al , 2006a,b) and the Brief Ataxia Rating Scale (BARS) (Schmahmann et al, 2009). Data collected from the validation trial in selected centers confirmed that the severity of A-T increased with age in patients under the age of 10 years, as demonstrated by a significant correlation of the total scores on the ICARS, SARA, and the BARS, but not for patients above 10 years. Overall, the distribution of scores for the ICARS for 5- to 10-year-old patients with A-T was similar to the scores for other validated scales such as the SARA, BARS, CGI-S (structured and unstructured versions). Please refer to Section 7.4 (Rationale for Efficacy Measures) for further details on the ICARS and RmICARS.

12.2 Clinical Global Impressions

The CGI (Guy, 1976) is the general name for 2 scales, the CGI - Change scale (CGI-C) and CGI- Severity scale (CGI-S). The CGI-S scale assesses the severity of the disease from 1 to 7 at a given point in time, while the CGI-C scale assesses the change in the participant's overall clinical condition from baseline using a 7-point scale, ranging from 1 (very much improved) to 7 (very much worse), with a score of 4 indicating no change.

The CGI-S evaluation will be performed at the Baseline Visit (videotaped), Visit 6, and Visit 9. The CGI-C will be assessed relative to the baseline assessment of the CGI-S. A video of the CGI-S interview and notes from the baseline assessment will be used to recall the baseline and anchor CGI-C to initial participant's status, to allow for a precise recall of participant's status by the rater. Given the comprehensive nature of the CGI-C assessment, all nuances of changes in participant's status will be captured.

To ensure independence of this rating from the rating of the ICARS (primary efficacy measure), a different rater will perform the CGI assessments. The rater for CGI must be a physician familiar with the participant but does not need to be a neurologist. The rater for CGI will be initially identified by the PIs based on experience and then qualified and trained by a specialized vendor on the administration of the CGI; will be blinded to the treatment, to safety assessments, and data; and will not have access to the ICARS scores. Neither the CGI nor ICARS raters will be allowed to administer trial treatment. Specific administration and scoring guidelines will be released to ensure consistency among raters and assessments.

For the CGI-C rating, clinicians will be required to conduct a full clinical interview and examination of the participant, if necessary, with the caregiver present. The interview and examination should assess various aspects of the participant's appearance (grooming, evidence of falls, etc.), neurological function, activities of daily living, cognition (orientation, calculation ability, language, ability to follow commands, memory, etc.), mood, and behavior.

12.3 Quality of Life Scale

The EQ-5D-Y descriptive system comprises the same 5 dimensions as the standard EQ-5D-5L, but uses more appropriate, child-friendly wording. The EQ-5D-Y consists of 3 pages: the title page, the EQ-5D-Y descriptive system (page 2), and the EQ VAS (page 3).

The 5 dimensions are: Mobility (walking about); Looking after myself; Doing usual activities; Having pain or discomfort; Feeling worried, sad, or unhappy. Each dimension has 3 levels: no problems/ no pain/not worried, some problems/some pain/a bit worried, a lot of problems/a lot of pain/very worried. Respondents are asked to indicate their own health state by checking the box next to the most appropriate response level for each of the 5 dimensions. Responses are coded as single-digit numbers expressing the severity level selected in each dimension.

The EQ VAS records the respondent's overall current health on a vertical VAS where the endpoints are labelled 'The best health you can imagine' and 'The worst health you can imagine'. The EQ VAS provides a quantitative measure of the respondent's perception of their overall health.

In the current trial, for participants aged 6 to 9 (included) years, the EQ-5D-Y Interviewer Administered Proxy version 1 will be used (i.e., a version of the questionnaire that is suitable for completion by a third party, e.g., a parent or caregiver, on the child's behalf). For participants 10 years of age and older, the self-reported version of the EQ-5D-Y is generally recommended.

The EQ-5D-Y administrator, proposed by the PIs, will be required to view the EQ-5D-Y didactic presentation and Patient Reported Outcome administration guidelines.

The EQ-5D-Y administrator, proposed by the PIs, will be required to view the EQ-5D-Y didactic presentation and patient-reported outcome administration guidelines.

12.4 Rater Requirements and Training

Properly qualified raters will need to be identified by each PI at each site to perform the ratings on the efficacy measures. The ICARS rater must remain blinded to other assessments, will not be involved in the rating of the CGI-S, CGI-C, QoL or C-SSRS, and will not have access to the safety data. The ICARS rater will not be allowed to administer the trial treatment. The ICARS ratings must be completed without consulting scores from the previous visit.

The rater for CGI will not have access to the ICARS ratings or safety assessments and data in scoring the CGI. The CGI rater will not be allowed to administer the trial treatment.

Qualification, training and certification process on the ICARS, CGI, EQ-5D-Y, and C-SSRS will be managed by a specialized vendor and will be described in specific guidelines. In addition, trainings sessions will be organized during Investigator's meeting and additional training and an intra-rater reliability (test-retest) assessment will be performed during the trial.

To maximize the consistency of the efficacy data, the same site's qualified, trained, and certified raters and with expertise in the field of AT disorders, will evaluate the same participant at approximately the same time throughout the trial.

12.5 Order of Test Performance

The ICARS should be the first scale administered, followed by the CGI-S/CGI-C and then by the EQ-5D-Y (please refer to Table 1 for the list of tests required at each Visit). Neurological assessments must be performed before any phlebotomy or IV insertion so that upper extremity neurological exam is not impeded by an IV line, except at screening and Visit 9.

13 SAFETY EVALUATIONS

The assessment of safety and tolerability will be based on the following:

a) Vital signs
b) Standard laboratory tests (clinical chemistry, hematology, and urinalysis)
c) 12-lead standard ECG
d) Physical and neurological examinations
e) Special laboratory parameters (CD4+ lymphocyte counts [a local laboratory test], alpha-fetoprotein, CRP)
f) Early morning plasma cortisol, ACTH (scheduled and as needed)
g) Hemolysis panel, as needed
h) Pregnancy testing
i) C-SSRS
j) BMD, where country regulations allow
k) Tanner staging
l) Sample sterility testing
m) Subjective reporting of any AE by the participant
n) Objective observation of any AE by the Investigator
o) The investigator will be asked to comment on any clinically significant abnormal test results.

The frequency of investigations that involve blood draws have been modified to ensure that the volume of blood taken from participants conforms to the guidelines specified for pediatric patients, as published by the World Health Organization (Howie, 2011).

13.1 Physical and Neurological Examinations

Physical and neurological examinations will be performed as specified on Table 1. The physical examination will include an examination of general appearance, skin, neck (including thyroid), eyes and ears, nose, mouth, throat, lungs, heart, abdomen, back, lymph nodes, extremities and nervous system. Genital, urinary tract, and rectal examinations are not required. The findings will be entered on the Physical Examination section (or AE, as applicable) of the eCRF. Clinically significant changes from Baseline should be assessed and should be recorded as an AE.

13.2 Vital Signs

Vital signs assessments will be performed as specified on Table 1 and will include body weight, temperature (method of recording to be documented in notes and eCRF), pulse, systolic and diastolic BP, and respiratory rate. Height will be measured by a stadiometer at Screening and each applicable subsequent visit and used along with body weight to calculate BMI. Pulse and BP will be measured after the participant has been in the supine position for at least 5 minutes. On the days of treatment, the vital signs will be measured within 15 minutes before and within 15 minutes after the IV infusion of the trial treatment. Weight and height will be measured just once per treatment visit and before the infusion.

Since there were no safety concerns from the vital signs analysis in the Phase 3 ATTeST trial, in the IEDAT-04-2022 trial, assessment of vital signs will not be performed in triplicate at each visit to avoid un-necessary burden to the trial staff and the participants. However, each investigator will be allowed to repeat measurements as needed in case previous attempts are not considered reliable or a value is abnormal.

Consistent with the ATTeST trial, in the IEDAT-04-2022 trial, vital signs will be taken pre and post treatment, to ensure participants' safety.

According to the usual practice in clinical settings, in case of an infusion of blood or derivates, the Sponsor recommends monitoring the participants enrolled in the trial in terms of heart rate and BP with the help of a surveillance monitor, including pulse oximeter. This recommendation should be followed during and after drawing of blood, as well as during infusion.

If a *change of clinical relevance* from pre-dose to post-dose is observed, the vital signs assessment should be repeated as often as needed, at the discretion of the Investigator and the observation period can be extended based on Investigator's judgement. Findings should be documented on the Vital Signs section (and AE section, if clinically significant according to Investigator's judgement) of the electronic CRF.

13.3 Electrocardiogram

All participants will have a standard 12-lead ECG performed as specified in Table 1. If clinically significant abnormal findings are noted at Screening and do not normalize on the repeat ECG evaluation at Baseline (done in triplicate), the patient will not be enrolled.

Review and interpretation of the ECG will be performed by a cardiologist or qualified physician at the investigational site, using a certified and serviced ECG machine.

Each ECG tracing must have the following information entered on it:

- Trial number
- Site number
- Participant's number and initials
- Date and time ECG obtained

If clinically significant abnormalities are found, the participant's ECG should be repeated at regular intervals until it returns to normal. The cardiologist or qualified physician reviewing the ECGs should use the following guidelines in determining the clinical significance of any abnormal findings:

- PR interval: <100 msec or >210 msec
- QRS interval: <50 msec or >120 msec
- QTc interval: >450 msec
- Heart rate: <50 bpm (sinus bradycardia) or >120 bpm (sinus tachycardia)
- Morphology: presence of T-wave inversion, abnormal R-waves, pathological Q-waves, or significant ST elevation or depression.

13.4 Standard Laboratory Evaluations and Screening Tests

Blood and urine samples for measurement of standard laboratory parameters will be as specified on Table 1. If clinically significant abnormal findings are noted at Screening and do not normalize on the repeat evaluation at Baseline, the participant will not be enrolled.

Evaluations of the hematology, clinical chemistry, and urinalysis analytes listed in Table 7 will be performed at each of the visits listed above. Details on aseptic procedure for blood withdrawal and on blood diversion are included in the "Aseptic Procedure Guideline" that will be provided to sites.

Table 7: Summary of Standard Laboratory Analytes

LABORATORY ANALYTES			
Hematology or Complete Blood Count	**Clinical Chemistry**		**Urinalysis (automated)**
Hematocrit	Sodium	Alkaline phosphatase	Color
Hemoglobin	Potassium	Lactate dehydrogenase	pH
RBC count	Chloride	creatine phosphokinase	Specific gravity
WBC count	Calcium	Triglycerides	Protein
Differential white blood cell count	Phosphorus	Total cholesterol	Glucose
• Neutrophils	Serum iron	HDL cholesterol	Ketones
• Lymphocytes	Bicarbonate	LDL cholesterol	RBC, WBC, casts *
• Monocytes	Glucose		Nitrites
• Eosinophils	Blood urea nitrogen		Bilirubin
• Basophils	Creatinine		Hemoglobin
Platelets	Total bilirubin		Urobilinogen
Mean corpuscular volume	Albumin		* Reflex microscopic analysis to be performed only if other analytes are abnormal on automated testing
Mean corpuscular hemoglobin	Total protein		
Mean corpuscular hemoglobin concentration	AST (SGOT)		
Red blood cell distribution width	ALT (SGPT)		

ALT = alanine aminotransferase; AST = aspartate aminotransferase; HDL = high-density lipoprotein LDL = low density lipoprotein; RBC = red blood cell count; SGOT = serum glutamic-oxaloacetic transaminase; SGPT = serum glutamic-pyruvic transaminase; WBC = white blood cell count

Special Diagnostic Tests: Female participants will have serum pregnancy test obtained at Screening. Females noted to be at Tanner Stage 2 or greater at Baseline will have a urine pregnancy test obtained and evaluated before each infusion at Visits 1, 4, 5, 6, 7, 8 and a serum pregnancy test at Visit 9/Early Discontinuation if noted to be at Tanner Stage 2 or greater at this Visit.

The Investigator must review screening laboratory values, as well as any repeat assessments, prior to the first administration of the trial treatment, to ensure that the participant meets the protocol's inclusion/exclusion criteria. The Investigator must review laboratory values from each subsequent evaluation within 24 hours of receipt of the laboratory report. After the review is completed, the Investigator must sign and date each laboratory report.

A central laboratory will be used for analyzing samples for routine and special laboratory tests, with the exception of ACTH and sterility test. Plasma cortisol will be tested centrally at screening and at V9/Early Withdrawal, unless for organizational/logistical/safety reasons the Investigator deems it preferable to test it locally. In certain circumstances the Investigator may choose to utilize a local laboratory if there is an immediate need for the test results.

The central laboratory will provide normal reference ranges for the laboratory tests on the laboratory results report. A value is considered normal when it falls on or within the upper and lower limits of the reference range for the laboratory. A value is considered abnormal when it exceeds the upper or lower limit of the reference range. The central laboratory will provide the normal reference ranges for each parameter and will verify that the result is not due to pre-analytical problems (e.g., sample taken improperly, sample stored incorrectly, sample labeled incorrectly) or to analytical problems (e.g., machine not accurately calibrated, technical problems with equipment or reagents, or deterioration of analyte).

The Investigator must evaluate any change of clinical relevance from pre-dose to post-dose in a laboratory test as to whether it meets the definition of an AE, and repeat, if needed, any clinically significant abnormal

laboratory test. Any laboratory abnormalities meeting the definition of an AE should be recorded on the AE CRF.

For the tests conducted at local level, the same applies but with reference to the normal reference ranges for the laboratory tests on the local laboratory results report.

Refer to Section 14, "Reporting Safety Information" for further instructions.

13.5 Special Laboratory Evaluations

Measurement of selected "special" laboratory parameters has been included in the trial to evaluate potential effects of eDSP treatment. The following special laboratory parameters will be assessed in the trial:

- CD4+ lymphocytes (a local laboratory test), α-fetoprotein (not repeated at baseline), CRP to be performed at Screening and Visits 1, 6, and 9.
- Screening for adrenal insufficiency will be performed in all participants, via early morning (before 8:00 AM) plasma cortisol testing, at the following times: (1) during the screening period (prior to randomization), (2) when participants are symptomatic, and (3) when participants are at most risk for adrenal insufficiency and adrenal crisis, such as following discontinuation or interruptions of dexamethasone dosing (i.e., loading failures and following discontinuation of the trial treatment). If the 8:00 AM cortisol level is below 3-5 µg/dL (depending on assay) regardless of symptoms, or the participant exhibits signs or symptoms of adrenal insufficiency (Appendix 7) and has a cortisol <10 µg/dL, the participant will receive a high-dose ACTH stimulation test as soon as possible. If the ACTH stimulation test is normal, the participant can be enrolled, or can continue dosing with EryDex, after the effects of the ACTH have dissipated (e.g., 30 days after ACTH administered, resulting in a prolongation of the Screening period). If the participant fails the ACTH stimulation test, they will be discontinued from trial treatment and from continuing further participation in the trial (not applicable at Visit 9) and referred to an endocrinologist (a pediatric endocrinologist, depending on the participant's age) with a recommendation to prescribe stress dose steroids.
- High-dose ACTH stimulation test – In the event that participants show signs or symptoms of adrenal insufficiency (Appendix 7) during the trial, especially following interruption of the trial treatment (including loading failures), they will be tested for adrenal insufficiency using a high-dose ACTH stimulation test (250 µg given IV or IM). If ACTH testing confirms adrenal insufficiency, the participant will be referred for evaluation and treatment. Blood samples (2.0 mL) for measurement of plasma cortisol will be collected prior to ACTH administration (0 minutes), and at 30- and 60-minutes post ACTH dose. A rise in plasma cortisol level to greater than 18 µg/dL within 60 minutes demonstrates a normal result. A rise in cortisol to less than 18 µg/dL within 60 minutes demonstrates an abnormal response.
- Results of plasma cortisol measurements and the ACTH stimulation test will be made available only to the PI and will not be available to raters performing the primary and secondary efficacy assessments.
- Hemolysis panel [free plasma hemoglobin, haptoglobin, bilirubin (total and conjugate), CBC, LDH and urinalysis] – to be performed as needed. See Appendix 4 for instructions on sample collection for these tests.

13.6 Optional Blood Samples for Biomarker Development

These samples will be collected, processed and stored according to instructions provided in the Laboratory Manual. They will be stored securely and labeled with a code that provides a link to the trial ID number for

each participant. Analyses will be performed by commercial and academic laboratories to identify useful biomarkers for the diagnosis and treatment of A-T.

13.7 Sterility Testing of EryDex

Before every EDS process, after the blood diversion, 1 mL of blood will be collected, for aerobic culture (see document "Aseptic Procedure Guideline" for further details).

[redacted]

13.8 Columbia-Suicide Severity Rating Scale

The C-SSRS (Posner et al, 2007; Posner et al, 2011) is a standardized suicidal rating system. Important psychometric properties and construct validity of the C-SSRS have been established in multicenter clinical trials.

Site personnel administering the C-SSRS will have been trained to use this assessment tool.

At Screening, an assessment of suicidality will be performed using the "Baseline/Screening" version of the C-SSRS. At Visit 1 and for all subsequent trial evaluations, assessments will be performed using the "Since Last Visit" version of the C-SSRS. Participants with suicidal ideation will be excluded from participating in the trial and referred to a mental health professional as needed.

13.9 Bone Mineral Density

Measurements of BMD will be performed, where country regulations allow, for all participants at Baseline and repeated at Visit 9 or Early Withdrawal to assess potential steroid-related changes. Bone mass will be measured in the spine and total body (less the head), following the guidelines provided in the 2013 International Society for Clinical Densitometry Official Pediatric Position (Gordon et al., 2014). The suggested method for assessing BMD in the trial is dual-energy x-ray absorptiometry. The analysis will be performed with Z-scores following the above guidelines. The same method of assessment will be used for each participant throughout the trial. BMD Z-scores will be evaluated according to the following chart:

BMD Z-Score	Level
- 1 or higher	Normal
Less than -1 to -2.4	Abnormal - osteopenia
-2.5 or lower	Abnormal - osteoporosis

Clinically significant changes from Baseline to Visit 9 should be documented as an AE or SAEs depending on the Investigator's judgement. Abnormal values at Baseline should be recorded as Medical History as they underline a pre-existing condition that frequently occurs in the A-T population.

13.10 Tanner Staging

Assessment of physical development, sexual maturation, and the effect of treatment with dexamethasone on these aspects will be evaluated using the Tanner scale (Marshall and Tanner 1969, 1970) starting at Screening, then as needed if there is evidence of changes in the development of a participant, and at Visit 9 or Early Withdrawal. The scale defines physical measurements of development in children, adolescents, and adults based on external primary and secondary sex characteristics, such as the size of the breast, genitals, testicular volume, and development of pubic hair. Due to natural variation, individuals pass through the Tanner stages at different rates, depending in particular on the timing of puberty. Tanner staging of the breasts in pre-menarchal females and of the scrotum in males who have not completed puberty will be performed.

Females who are noted to reach Tanner Stage 2 at the Baseline or display any evidence of childbearing potential will undergo pregnancy testing prior to trial treatment administration and serum pregnancy test at Visit 9/Early Discontinuation, if noted to be at Tanner Stage 2 or greater at this Visit. Should the PI/delegated Investigator note evidence, or should the parent report any evidence of female trial participants approaching puberty (e.g., breast development, pubic hair, or onset of menses) after the Baseline and during the trial, a full Tanner staging will be performed to ensure female participants will start pre-treatment pregnancy testing, as needed.

14 REPORTING SAFETY INFORMATION

14.1 Adverse Events

Assessment of AEs will be performed throughout the trial, from the time of signing of the ICF at the start of the Screening period through the final trial visit (Visit 9). All AEs will be recorded in the eCRF.

14.1.1 *Glossary*

14.1.1.1 Adverse Drug Reaction

In the pre-approval clinical experience with a new medicinal product or its new usages, particularly as the therapeutic dose(s) may not be established, all noxious and unintended responses to a medicinal product related to any dose should be considered adverse drug reactions. The phrase “response to a medicinal product” means that a causal relationship between a medicinal product and an AE is at least a reasonable possibility (i.e., the relationship cannot be ruled out).

14.1.1.2 Adverse Event

An AE is any untoward medical occurrence in a participant or clinical investigation participant administered a pharmaceutical product and that does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

14.1.1.3 Serious Adverse Event or Serious Adverse Drug Reaction

Any untoward medical occurrence that at any dose:

- Results in death,
- Is life-threatening, i.e., an event which, in the view of the Investigator, places the participant at immediate risk of death from the event as it occurred (it does not include an event which hypothetically might have caused death if it were more severe)
- Requires inpatient hospitalization or prolongation of existing hospitalization,
- Results in persistent or significant disability/incapacity,
- Is a congenital anomaly/birth defect (if exposure to product just before conception or during pregnancy resulted in an adverse outcome in the child), or
- Is an important medical event that may jeopardize the participant or may require medical intervention to prevent one of the outcomes listed above.

Symptoms or medically significant laboratory or instrumental (e.g., electrocardiographic) abnormalities of a pre-existing disease, such as cancer or other disease, should not be considered an AE. However, the occurrence of new symptoms, or laboratory or instrumental abnormalities, as well as worsening of pre-existing symptoms, are considered AEs.

14.1.1.4 Unexpected Adverse Drug Reaction

An Unexpected Adverse Drug Reaction is an adverse drug reaction, the nature or severity of which is not consistent with the applicable product information (e.g., IB for an unapproved investigational product or Package Insert/Summary of Product Characteristics for an approved product).

14.1.1.5 Non-Serious Adverse Event

A non-serious AE is any AE that does not meet the criteria listed above for an SAE.

14.1.2 *Data Collection*

For each event, record the following information on the AE section of the CRF:

- **Classification of the Event:** Classify the event as either serious or non-serious
- **Description of Signs or Symptoms:** Whenever possible, record a specific diagnosis for the event. If a diagnosis cannot be made, then record each sign or symptom separately (e.g., record nausea and vomiting as 2 events). If multiple episodes of an event occur, separated by an appropriate time interval to justify considering the subsequent episodes as a repeat occurrence, record each episode separately on the CRF
- **Onset Date and Time:** Record the date and time the event started. If a change from baseline/previous evaluation in a laboratory test is reported as an AE, record the start date as the date of collection of the first lab sample that shows the change
- **Stop Date and Time:** Record the date and time the event resolved. If a change from baseline/previous evaluation in a laboratory test is reported as an AE, record the stop date as the date of collection of the first sample that shows a return to the previous level
- **Intensity:**
  1. Mild: Event not resulting in disability/incapacity, which resolves without treatment.
  2. Moderate: Event not resulting in disability/incapacity, which requires treatment.
  3. Severe: Event resulting in temporary and mild disability/incapacity, which requires treatment.

- **Relationship to the Trial Agent:** Every effort should be made to determine the cause of each AE. The correlation between the trial treatment and the AE should be classified as follows:

Probably Related	The event follows a reasonable temporal sequence from administration of the trial treatment; The event follows a known response pattern to the trial treatment; The event cannot be reasonably explained by: • the known characteristics of participant's clinical state, or • by other therapy administered, or • by the diagnostic/interventional procedure. There is evidence of partial or complete disappearance of the event after withdrawal of the product (positive de-challenge)
Possibly Related	The event follows a reasonable temporal sequence from administration of the trial treatment; Causation of the event by the trial treatment cannot be excluded; The event follows a known response pattern to the trial treatment but the event could have been produced by: • the participant's clinical state, or • other therapy administered, or • a diagnostic/interventional procedure.
Unlikely Related	The adverse event follows a reasonable temporal sequence from administration of the trial treatment; Other reasons are more likely to be the cause of the adverse event, based on the present knowledge of the • disease under treatment, or • other therapy administered, or • trial treatment. A causal relationship between the adverse event and the trial treatment cannot be ruled out with certainty.
Not Related	The event is either a pre-dose event or is definitely due to causes separate from the administration of the trial treatment, i.e., • documented pre-existing condition • technical and/or manual procedural problems • concomitant medication • participant's clinical state

Action Taken, in relation to treatment of the AE:

0. None
1. Drug treatment required (a medication was prescribed or changed; record on the Concomitant Medication section of the CRF)
2. Non-drug treatment required (a non-drug treatment was prescribed or changed, record under "Comments" in the AE section of the CRF)
3. Hospitalization or prolonged hospitalization (fill out an SAE report)
4. Diagnostic or clinical test(s) conducted (attach a copy of the results to the CRF)
5. Participant discontinued from the trial

Action taken with trial treatment

0. Dose not changed
1. Treatment interrupted
2. Treatment withdrawn
3. Not applicable

Participant Outcome:

1. Recovered without sequelae
2. Recovered with sequelae (describe the sequelae under "Comments" in the AE section of the CRF)
3. Not Recovered, event on-going (follow the participant until a definite outcome can be determined. When follow-up data are collected, report follow-up information under "Comments" in the AE section of the CRF; if event is serious, fill in a follow-up SAE Report.)
4. Died (list primary cause of death under "Event Description" of the AE section of the CRF; if available, attach a copy of the autopsy report to the CRF and send a copy to the Sponsor)

Comments:

Provide other pertinent clinical information and observations under "Comments" in the AE section of the CRF. For example, record predisposing or contributing conditions, such as previous history, concomitant diseases or medications, and/or procedural risks.

Events of positive EryDex sample of culture-based sterility test results should be generally reported as AEs, representing an abnormal laboratory finding, unless a participant shows sign or symptoms of blood borne infection (in this case, the event may be reported as an SAE, depending upon whether it meets that definition). Detailed instructions on the reporting of such cases will be provided as part of the training for the Investigators.

14.1.3 *Participant Follow-up*

Every attempt should be made to follow the participant until the AE has resolved or until the Investigator determines the participant has returned to an acceptable state of health.

14.1.4 *Reporting Serious Adverse Events*

The Investigator must **report all SAEs within 24 hours of event awareness**, irrespective of the relationship to trial treatment, to the CRO pharmacovigilance department, by completing the dedicated section within the electronic data capture (EDC) system, which will then be automatically sent by the EDC system to the pharmacovigilance department e-mail. Alternatively, if the EDC is not functional, the investigator may complete the appropriate reporting form and send notification by e-mail at the following address.

PVDS-ROW@premier-research.com

The CRO pharmacovigilance department will then forward this information to Quince Therapeutics S.p.A within 1 business day of receipt. The contact details and e-mail address of the CRO pharmacovigilance department (including back-up contacts) will be communicated to the investigators by the CRO before or during Site Initiation Visits.

The minimum information required for an initial report of an SAE is as follows:

- Sender of report (name, address of Investigator, site number),
- Participant identification (screening number),
- Protocol number,
- Reportable event.

In case of death, a comprehensive narrative report of the case should be prepared by the Investigator and sent to the CRO pharmacovigilance department by e-mail together with the Serious AE Form, retaining a copy on-site. If an autopsy is performed, a copy of the autopsy report should be actively sought by the

Investigator and sent to the CRO pharmacovigilance department as soon as available. A copy of the autopsy report will be retained on-site.

A follow-up SAE Report will be completed by the Investigator if important follow-up information (i.e., diagnosis, outcome, causality assessment, results of specific investigations) are made available after submission of the initial form. The **follow-up form** will be sent to the CRO pharmacovigilance department as described above and **within 24 hours of follow-up information awareness**.

If the Investigator becomes aware of any deaths or SAEs suspected of being causally related to the trial treatment after the end of the window established in the protocol following investigational product administration, they will be reported to the CRO pharmacovigilance department as described above, unless a participant has been already enrolled in the OLE trial at the time of event occurrence (in this case, the SAE would be reported in the context of the OLE trial).

14.2 Reporting Events Concerning the Medical Device

Although events concerning the medical device components of the EDS do not necessarily impact participants' safety, Investigators are required to inform Quince Therapeutics S.p.A of any serious or non-serious incident and any device-related AEs and product deficiencies concerning Quince Therapeutics S.p.A medical devices, through the Medical Device Report Log. Information should be sent to *technicalservice@quincetx.com* immediately and not later than 12 hours from the event.

Quince Therapeutics S.p.A is responsible to handle and report such device events according to applicable country specific regulatory requirements.

14.3 Adverse Events of Special Interest

The following TEAEs are considered to be adverse events of special interest (AESI) due to their potential relationship to the trial treatment:

- **Adrenal insufficiency:** New onset of adrenal insufficiency (based on clinical symptoms and confirmed with either low cortisol and/or an abnormal ACTH stimulation test).
- **Infections of special interest:** New onset bacteremia, sepsis, pneumonia, or opportunistic infections (e.g., candida sepsis, pneumocystis pneumonia, tuberculosis, toxoplasmosis, varicella zoster virus infection, herpes ophthalmicus, cytomegalovirus infection, aspergillosis, histoplasmosis).
- **Iron deficiency anemia:** New onset of iron deficiency anemia supported by laboratory testing (low hemoglobin and another measure suggesting low iron levels [e.g., low ferritin level, low serum iron, high total iron-binding capacity, low iron saturation]). Although iron deficiency anemia is typically mild and does not require rapid communication, this AE was selected as being of special interest since treatment with EryDex requires monthly blood draws in children who often have an underlying iron deficiency.

14.4 Safety Reporting to Investigators, Institutional Review Boards/ Independent Ethics Committees, and Regulatory Authorities

The Sponsor or their designee will be responsible for reporting all SAEs to regulatory authorities, investigators, and Independent Ethics Committees (IECs) / Institutional Review Boards (IRBs), as applicable, in accordance with national regulations for all the involved countries. The Sponsor or their designee will prepare and distribute expedited safety reports according to all applicable laws and regulations. The investigational site also will forward a copy of all expedited reports to their IRB/IEC.

14.5 Reporting Suspected Unexpected Serious Adverse Reactions (SUSAR)

A SUSAR is defined as an untoward and unintended response to a study drug, which is not listed in the applicable product information, has a reasonable possibility of a causal relationship and meets one of the following serious criteria: results in death, is life-threatening, requires hospitalization or prolongation of an existing hospitalization, results in persistent or significant disability or incapacity, or is a congenital anomaly or birth defect.
When an SAE report form is received and evaluated by Sponsor or designee and this event is considered Unexpected and possibly related to the investigational product (therefore qualified as SUSAR), a report must be submitted to all applicable national and local authorities according to national and local laws:

- Within 7 calendar days of the Sponsor's (or designated CRO's) awareness if fatal or life-threatening
- Within 15 calendar days of the Sponsor's (or designated CRO's) awareness for non-fatal or non-life threatening SUSARs

14.6 Reporting Overdose

If the investigational site staff administering the trial medication reports that a participant was given more than the specified dose of trial medication, this will be considered an overdose and must be reported immediately to the Investigator. Any instance of overdose, whether symptomatic or not, must be reported using a process similar to SAE reporting. Details of any signs or symptoms and their management should be recorded, including details of any antidote(s) administered.

14.7 Pregnancy

This trial will exclude pregnant and breastfeeding participants; females of childbearing potential will be excluded, unless practicing adequate contraception, as determined by their Healthcare Provider. Further precautions will be taken for females at Tanner Stage 2 or greater. Female participants will have serum pregnancy tests obtained at Screening and then females noted to be at Tanner Stage 2 or greater at Baseline will have urine pregnancy tests obtained and evaluated before each infusion at Visits 4, 5, 6, 7, and 8 and a serum pregnancy test at Visit 9/Early Withdrawal if noted to be at Tanner Stage 2 or greater at that visit.

Following the recommendation of the Heads of Medicines Agencies / Clinical Trials Facilitation and coordination Group (Recommendations related to contraception and pregnancy testing in clinical trials; Version 1.1, 21 Sept 2020), adequate birth control methods (i.e., methods that can achieve a failure rate of less than 1% per year when used consistently and correctly) include:

- Combined (estrogen and progestogen containing) hormonal contraception associated with inhibition of ovulation:
  - Oral
  - Intravaginal
  - Transdermal
- Progestogen-only hormonal contraception associated with inhibition of ovulation
  - Oral
  - Injectable
  - Implantable
- Intrauterine device
- Intrauterine hormone-releasing system
- Bilateral tubal occlusion

- Vasectomized partner
- Sexual abstinence

If a participant becomes pregnant during the trial, she will be discontinued from the trial treatment and trial immediately. Participants and their parents/caregivers will be instructed to notify the Investigator if it is determined that, after completion of the trial they have become pregnant, either during the treatment phase of the trial or within 28 days (-7/+2)days of completing the trial treatment.

Whenever possible a pregnancy should be followed to term, any premature terminations reported, and the status of the mother and child should be reported to the CRO or Quince Therapeutics S.p.A after delivery.

14.8 Breaking the Trial Blind by the Investigator

In case of emergency, the Investigator may break the blind for an individual participant, if the knowledge of the administered compound is deemed necessary for the safety of the participant Breaking of the blind will be performed by the Investigator by calling/logging into the IWRS system.

If the trial blind is broken, the Investigator must immediately notify the CRO and provide the participant's number. The reason for breaking the blind should be noted on the AE (or SAE) section of the CRF. Quince Therapeutics S.p.A will inform the Independent Safety Monitoring Board of the unblinding.

14.9 Independent Data Safety Monitoring Board

An iDSMB will be established by Quince Therapeutics S.p.A to review the safety of all participants enrolled in this trial, on an ongoing basis. No Quince Therapeutics S.p.A employee or investigator involved in the Quince Therapeutics S.p.A clinical studies will be a voting member of this board. The iDSMB will regularly review the safety data as it accrues.

The Board will be regularly notified of the occurrence of any unexpected and at least possibly related fatal or life-threatening events immediately (within 7 calendar days) and any other unexpected and at least possibly related SAEs within 15 calendar days. The Board will also receive updates on any discontinuations due to AEs on a regular basis. The Board will have access to the unblinded safety data including SAEs and discontinuations due to AEs, as well as clinically significant abnormal laboratory tests, vital signs and ECGs at periodic intervals.

The iDSMB will review all of the safety data on an ongoing basis, with special emphasis on the incidence and severity of steroid-related events, new infections, and SAEs and deaths, in addition to the standard safety parameters. After reviewing the emerging safety profile for EryDex, the Board will make a recommendation to Quince Therapeutics S.p.A to (a) amend the ongoing trial (e.g., increase safety monitoring, modify dosing), (b) terminate the EryDex program (e.g., the EryDex safety profile is unacceptable), (c) continue the clinical program as designed.

The iDSMB charter (separate document) will be submitted to regulatory authorities, as needed, and will be available to IECs/IRBs upon request.

15 PARTICIPANT COMPLETION AND DISCONTINUATION

15.1 Definitions

A participant will be considered to have 'completed' the trial when he/she returns for the final evaluations at Visit 9. 'Discontinuation' will refer to any participant who does not complete the full treatment period of the trial. In the context of the IEDAT-04-2022 trial, the permanent withdrawal of the trial medication aligns to the discontinuation from the trial.

15.2 Procedures for Handling Withdrawals

In the absence of a medical contraindication or significant protocol violation, every effort should be made by the Investigator to keep the participant in the trial. However, should the participant be withdrawn before Visit 9, all efforts should be made to complete all final evaluations and report the observations as thoroughly as possible at the time of the participant's withdrawal, with an explanation of why the participant is withdrawing from the trial.

The criteria for discharging a participant from the trial, including receiving trial treatment, prior to the final trial examination are listed below. The Investigator must indicate the primary reason (only one can be reported) for discontinuation, as well as the date when the decision was made; these will be specified on the 'End of Trial' eCRF form.

A participant may be withdrawn from receiving trial treatment and therefore from trial participation if:

- any hypersensitivity or allergic reaction, clearly linked to the trial medication, has occurred;
- the participant experiences an AE sufficiently severe, in the opinion of the investigator, that it contraindicates the participant continuing to receive the trial treatment and the participating in the trial; if the participant experiences a systemic illness considered unrelated to the trial medication, it still must be reported as an AE;
- if participants have CD4+ lymphocyte counts consistently below 150/mm^3 (for participants ≥7 years old), or below 200/mm^3 (for participants ≥7 years old and in presence of oral infections, like oral candidiasis, documented at screening or recurrent as per medical history/AEs documentation) or below 400/mm^3 (for participants <7 years old) during treatment, the PI should contact the Medical Monitor to discuss if it may be necessary to interrupt trial treatment;
- participants failing the ACTH stimulation test will be discontinued from the trial and referred to an endocrinologist (pediatric endocrinologist, depending on the participant's age) with a recommendation to prescribe stress dose steroids;
- the participant/caregiver withdraws consent (e.g., participant refuses to have any more blood samples taken for the EryDex process; in this instance a specific reason must be recorded by the investigator);
- the participant becomes pregnant, or a female of childbearing potential does not agree to undergo pregnancy testing or use an adequate birth control method;
- a major protocol deviation that jeopardizes the continued well-being of the participant or poses a risk to the participant's health; if the participant is able to abstain in the future from the activities/behaviors that constituted a major protocol violation, the participant should be allowed to continue;
- the participant is lost to follow-up, i.e., the participant did not return to the clinic, and attempts to contact the participant were unsuccessful. Attempts to contact the subject must be documented
- the Sponsor, IEC/IRB), or regulatory agency terminates the trial.

Dropouts will not be replaced.

The Investigator may terminate a participant's trial participation at any time during the trial if a participant meets the trial treatment/trial termination criteria described above. In addition, a participant or their parent/guardian may discontinue the participant's participation without giving a reason at any time during the trial. Should a participant's participation be discontinued, the primary reason for termination must be recorded. For participants who discontinue prior to the completion of the full Treatment Period, an attempt should be made to perform all Trial Completion (Visit 9) assessments on the participant, and to follow up on any safety issues until resolution. Participants who discontinue prematurely, but return for scheduled

efficacy evaluations, will become part of the retrieved dropout (i.e., participants who discontinue treatment or dropout and return for the final visit) analysis population.

Participants who complete the full treatment period, complete the trial assessments, and provide informed consent will be eligible for entry into an OLE trial, provided there are no issues that would preclude their continuation in the trial.

Participants who discontinue prematurely will still be required to perform the Trial Completion visit (Visit 9).

16 STATISTICAL METHODS

16.1 Analysis of Efficacy

16.1.1 *Primary Efficacy Endpoint*

To evaluate the effect of EryDex on CNS symptoms, as measured by the change of the RmICARS from baseline to Visit 9 compared to placebo in A-T (6- to 9-year-old participants primary analysis population). The RmICARS is a re-scored version of the ICARS and consists of 9 items across 3 domains (kinetic domain and speech limited to one item each), totaling a maximum score of 29 points.

16.1.2 *Key Secondary Efficacy Endpoint*

To evaluate the overall clinical effect of EryDex, compared to placebo, in A-T (6- to 9-year-old participants primary analysis population), based on:

- CGI-S from baseline to Visit 9.

16.1.3 *Other Secondary Efficacy Endpoint*

- CGI-C from baseline to Visit 9.

16.1.4 *Exploratory Endpoints*

- Health-related QoL using the change from baseline to Visit 9 of the EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older
- Health-related QoL using the change from baseline to Visit 9 of the EQ-5D-Y total score and VAS

16.1.5 *Participants Aged 10 Years and Older*

These primary/secondary/exploratory efficacy objectives will also be evaluated for participants aged 10 years and older. These analyses will be performed as data summaries in this older age cohort.

16.2 Statistical Methods

16.2.1 *Sample Size*

The sample size for this trial is based on a comparison between active and control in the change from baseline to Visit 9 for the RmICARS. The null hypothesis will be no difference in the change from baseline RmICARS between active and control, with the alternative hypothesis of a difference of -2.4 between active and control (ATTeST trial Clinical Trial Report). A total sample size of 86 participants 6 to 9 years old (43 per treatment group) will provide approximately 90% power with a 2-sided test at 0.05 type I error, assuming a SD of 3.0 for the treatment group and 3.7 for the control group.

The number of participants aged 10 years and older is not based on the ability to detect a specific treatment effect. These participants will not be included in the main analysis population for efficacy.

16.2.2 *Populations for Analysis*

The following analysis sets will be used:

- *Intent-to-treat (ITT) Population*: the ITT Population is defined as all participants who were randomized.
- *ITT (6-9) Population:* the ITT (6-9) Population is based on the ITT Population and comprises randomized participants aged 6 to 9 years. This population will be the one on which main efficacy analyses are conducted.
- *ITT (10+) Population:* the ITT (10+) Population comprises randomized participants aged 10 years and older.
- *Per-Protocol (PP) Population:* a subset of the ITT (6-9) Population comprising all participants aged 6 to 9 years who qualified for the ITT (6-9) Population, met trial treatment adherence requirements, completed Visit 9, and did not have disqualifying protocol deviations as specified in the SAP. Classification of participants in the PP Population will be done in a blinded manner before database lock.
- Safety Population (SP): any participant who received at least 1 dose of the randomized treatment.

16.2.3 *Background and Demographic Characteristics*

The background and demographic characteristics will consist of age, sex, race, ethnicity, height, body weight, BMI, past and current medical conditions, history of disease, and genetic confirmation of A-T. Continuous variables will be summarized by mean, SD, median, and range (minimum, maximum), and discrete variables will be summarized using frequencies and percentages.

16.2.4 *Trial Medication*

The number of participants receiving each dosing condition (DSP or placebo) will be reported, and the average dose of EryDex (DSP encapsulated in autologous erythrocytes) in participants administered the active treatment (Group 1, Safety population), based on measurements of samples taken from the infusion bags prior to dosing, will be summarized by mean, SD, median, and range (minimum, maximum).

16.2.5 *Concomitant Medications and Therapy*

A listing of concomitant medications administered from the time of dosing of the trial medication through completion of the final evaluation (Visit 9 or early discontinuation) will be provided by treatment group. Concomitant medication taken during the Screening period will be listed separately.

16.2.6 *Safety Evaluations*

All participants in the SP will be included in the safety analyses. All AEs will be listed and summarized by body system and preferred term. The incidence of AEs (%) and their intensity and relatedness to the trial treatment, as assessed by the Investigator, will be reported. SAEs and events which are newly occurring or worsening after administration of the trial treatment will be summarized. In addition, AEs that result in death or discontinuation of the trial treatment will be listed separately. All AEs experienced after receiving treatment will be considered as TEAEs.

Other safety parameters such as vital signs, laboratory parameters, ECGs, and physical/neurological examination findings will be listed and summarized accordingly, including their changes and summaries of clinically significant changes. Abnormal and clinically notable values will be identified and listed for each parameter, as appropriate. Where feasible, mean changes from baseline will be examined using a paired t-test.

Safety analysis will be performed on the total population, on the 6- to 9-year-old participants, and on participants ≥10 years old.

16.2.7 *Efficacy Analyses*

The full statistical analysis details will be provided in a SAP prior to database lock and unblinding of the trial.

16.2.7.1 Handling of Dropouts and Missing Data

Participants who discontinue treatment prematurely will be encouraged to return for assessments at Visit 9. A hypothetical strategy will be used to address participants who are not able to provide a Visit 9 assessment due to termination of treatment or otherwise lost to follow-up. Missing data will be handled as described in the analysis of the primary efficacy parameter. Retrieved dropouts will be used in sensitivity analyses for the primary endpoint. Sensitivity analyses for missing data will be specified for each endpoint in more detail in the SAP.

16.2.7.2 Hypothesis Testing and Handling of Multiple Comparisons

The primary efficacy endpoint will be declared statistically significant at alpha 0.05 two-sided. A hierarchical testing procedure will be used to control for multiplicity to maintain overall alpha at 0.05. Specifically, CGI-S will be tested only if the test of RmICARS resulted in $p < 0.05$, and the CGI-C would be tested only if CGI-S $p < 0.05$.

16.2.7.3 Primary Efficacy Endpoint and Analysis of the Primary Efficacy Parameter

The primary efficacy endpoint, the change from baseline to Visit 9 in RmICARS in participants aged 6 to 9 years old, will be analyzed using a mixed-model repeated-measures analysis, as it allows for data missing at random to be imputed from this model. Data missing for reasons other than treatment discontinuation due to AEs or deaths will be imputed this way. The analysis will be conducted by imputing datapoints after death to be the worst value for the endpoint at that visit across the trial, and by imputing datapoints after treatment discontinuation due to AEs using a Jump to Reference approach which assumes the RmICARS score behavior for participants withdrawing due to AEs is the same as in the control group. The analysis model will include the change from baseline to Visits 6 and 9 as the repeated measure, with treatment as the fixed effects and terms for region, sex, and visit, and baseline RmICARS, sex, and age as continuous covariates, and the treatment-by-visit interaction. The treatment effect, together with the associated 2-sided 95% CI and p-value will be calculated. Unstructured covariance matrixes will be used. Model effect estimation will be based on restricted maximum likelihood. Sensitivity analysis using a retrieved dropout imputation method for discontinuation due to AEs will be performed to address possible issues related to missing data.

The null (H_0) and alternative (H_1) hypotheses for the efficacy analysis of change from baseline in the RmICARS total score are:

H_0: There is no difference between Group 1 (DSP) and Group 2 (Placebo) with respect to RmICARS;

H_1: There is a difference between Group 1 (DSP) and Group 2 (Placebo) with respect to RmICARS.

16.2.7.4 Secondary Efficacy Endpoints

The change in CGI-S will be considered the key secondary trial outcome. The change in CGI-S from baseline to Visit 9 will be classified as improved versus no change/worsened. The proportion improved will be analyzed with logistic regression, with covariates for age, baseline RmICARS, sex, and region.

CGI-C: the difference in the outcome on this ordinal scale at Visit 9 between treatment groups will be analyzed with a Proportional Odds logistic regression model. The resulting proportional odds ratio represents the likelihood of participants in the EryDex group being in a better level of the CGI-C compared to the participants in the placebo group. The proportional odds model will include covariates for baseline RmICARS, age, sex, and region. Summary statistics will be used to show the proportion of participants in each level, as well as the individual odds ratios at each level. Should a level contain fewer than 5 participants across treatments, or only 1 participant in either treatment arm, the level will be collapsed to the next higher (worse) level. Within the range of responses, the proportion of participants who report any improvement (levels 1-3, inclusive) versus those who report no change or worsening (levels 4-7, inclusive) will be of primary interest.

If the proportional odds assumption is violated, then a logistic regression model with covariates for age, baseline RmICARS, sex, and region will be used to compare the 2 treatment arms. The response variable for this logistic regression will be the proportion of participants who report improvement (Levels 1-3) versus no change and worsened (Levels 4-7).

16.2.7.5 Exploratory Endpoints

The exploratory endpoint is the EQ-5D-Y Interviewer Administered Proxy version 1 for participants 6 to 9 years of age and self-reported for participants 10 years of age and older with each of the 5 domains evaluated, and a total score calculated. VAS will be reported separately on a scale from 0 to 100. The change from baseline to Visit 9 in total score and VAS will be analyzed with Analysis of Covariance, with baseline score, region, sex, baseline RmICARS, and age as covariates.

As a supplemental analysis, the proportion of participants who do not worsen in total score and VAS will be compared between the 2 treatment arms with logistic regression. The logistic regression model will include covariates for baseline score, region, sex, baseline RmICARS, and age.

16.2.7.6 Participants Aged 10 Years and Older:

These primary, secondary, and exploratory efficacy objectives will also be evaluated for the participants aged 10 years and older. These analyses will be performed as data summaries in this older age cohort.

17 ETHICS

17.1 Ethical Considerations

The trial will be carried out in accordance with the Declaration of Helsinki, as amended by the 64th General Assembly of the World Medical Association, Fortaleza, Brazil, October 2013.

In addition, this trial will be conducted in accordance with the following:

- European directive 2005/28/CE dated 8 April 2005 (GCP) setting the GCP for biomedical research on drugs for human use.
- Regulation (EU) 2016/679 of the European Parliament and of the Council of 27 April 2016 on the protection of natural persons with regard to the processing of personal data and on the free movement of such data.
- Regulation (EU) No 536/2014 of the European Parliament and of the Council of 16 April 2014 on clinical trials on medicinal products for human use, and repealing Directive 2001/20/EC.

17.1.1 *Participant Information and Informed Consent*

The ICF, as well as the Participant Information Sheet, must be approved by the IRB/IEC together with the Trial Protocol before the start of the trial.

For all participants, their parent/caregiver or legal representative (with assent by the participant), must sign and personally date an approved ICF after receiving detailed written and verbal information about the reason, the nature, the required procedures, the intended duration, and the possible risks and benefits and any discomfort associated with the trial. Each participant/parent/caregiver should be informed that participation in the trial is voluntary and that he/she may refuse to participate or withdraw from the trial at any time, without penalty or loss of benefits to which the participant is otherwise entitled.

The language used in the oral and written information about the trial, including the written ICF, should be as non-technical as practical and should be understandable to the participant. The participant must be given ample time to read and to understand the Participant Information Sheet and opportunity to inquire and ask any clarification about the trial before signing the ICF.

No trial procedure can be performed (including the screening visit) before the informed consent form has been signed. The informed consent procedure must be done according to the guidelines provided in the Declaration of Helsinki and the International Conference for Harmonisation (ICH) E6 Guideline for Good Clinical Practices (GCP).

The participant/parent/caregiver must be made aware and agree that personal information may be scrutinized during audit/inspection by competent authorities and properly authorized persons. However, personal information will be treated as strictly confidential and will not be publicly available.

By signing the Investigator Statement (Appendix 1), the Investigator assures Quince Therapeutics S.p.A that Informed Consent will be obtained.

Original signed ICF s will be filed with the Investigator's File.

17.1.2 *Notification*

The Sponsor may submit different types of notification in the Clinical Trial Information System (CTIS) as serious breach: this allows the Sponsor to inform about a breach likely to affect to a significant degree the

safety and rights of a volunteer or the reliability and robustness of the data generated in the Clinical Trial (CT). These notifications must be made without undue delay but no later than 7 days from the date on which the Sponsor became aware of the breach (article 52 of the CT Regulation).

In case of Personal Data Breach, the Sponsor (i.e., Controller) informs the Supervisory Authority without undue delay and where feasible, or later than 72 hours after becoming aware of it. When the notification is not made within 72 hours, reasons for delay are added to the notification.

17.1.3 *Data Protection*

This trial will be performed in accordance with the standard operating procedures of the Sponsor (or designee), GCP, the EU Clinical Trials Directive and the Code of Federal Regulations (CFR), the guidelines of the International Council for Harmonisation (ICH), any applicable local regulation, and the most recent guidelines of the Declaration of Helsinki.

All personal data will be processed according to Regulation (EU) 2016/679 on General Protection of personal Data (RGPD), and as per local data protection regulations.

The Investigator must guarantee the confidentiality of the trial data in the medical files by implementing security measures to prevent unauthorized access to the data and to the computer system.

- Participants will be assigned a unique identifier by the Sponsor. Any participant records or datasets that are transferred to the Sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred. The participant must be informed that their personal trial-related data will be used by the Sponsor in accordance with local data protection law. The level of disclosure must also be explained to the participant.
- The participant must be informed that their medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the Sponsor, by appropriate EC members, and by inspectors from regulatory authorities.

17.2 IEC/IRB Approval

The protocol, Investigator's Brochure, , Participant Information Sheet, ICF, and any advertisement for the recruitment of participants must be reviewed and approved by an appropriately constituted IEC/IRB, as required in Chapter 3 of the ICH E6 Guideline. A copy of the Committee's dated approval and a list of the members of the IEC/IRB will be given to the Sponsor for the Sponsor's files. A copy will be also included in the Final Report. Written IEC/IRB approval must be obtained by the Sponsor prior to shipment of trial treatment or participant enrollment. Any amendments to the protocol, ICF, or Participant Information Sheet, other than administrative ones, must be approved by this committee.

18 ADMINISTRATIVE CONSIDERATIONS

18.1 Regulatory Requirements: Sponsor/Investigator Obligations

This trial will be conducted in accordance with the Declaration of Helsinki and the ICH E6(R2) Guideline (GCP). To ensure compliance the Investigator agrees, by written consent to this protocol, to fully cooperate with compliance checks by allowing access to all documentation, including participants' hospital files (the source documents), by authorized individuals.

18.2 Curriculum Vitae

The Investigator and any sub-investigator(s) must provide the Sponsor with current copies of their own curriculum vitae.

18.3 Investigator and Trial Administrative Structure

The administrative structure of the trial (e.g., Investigators, monitoring and evaluation personnel, laboratory facilities, clinical trial supply management) is presented in the Trial Master File.

The listing should include:

- The Investigator(s);
- Any other person carrying out observations of primary or other major efficacy or safety variables, such as a nurse, physician's assistant, clinical psychologist, clinical pharmacist or house staff physician.

18.4 Investigator's Statement

This document, signed and dated by the PI, describes the Investigator's obligations. The standard text is appended to the protocol (Appendix 1).

18.5 Monitoring Procedures

18.5.1 *Trial Monitoring*

A CRO will be selected by the Sponsor to oversee the conduct of the trial. An appropriate representative of the CRO (Trial Monitor) will maintain contact with the Investigator and will visit the trial site for the purpose of discussing and/or retrieving data.

An initiation (pre-study) visit will be made by the Study Monitor to discuss with the Investigator the protocol and the obligations of both the Sponsor and the Investigator. The Investigator must allow the trial monitor to perform periodic, interim monitoring visits.

The purposes of these visits are:

- To verify that written Informed Consent was obtained prior to each participant's participation in the trial
- To assess the progress of the trial
- To review the compliance with the trial protocol
- To determine whether all AEs were appropriately reported
- To determine whether the Investigator is maintaining the essential documents
- To discuss any emergent problem
- To check the CRFs for legibility, accuracy and completeness

- To validate the contents of the CRFs against source documents
- To collect completed CRFs
- To assess the status of drug storage, dispensing and retrieval

The Investigator will make available the source documents for inspection. This information will be considered as confidential. *Violations of and deviations from the protocol must be notified to the Trial Monitor as soon as possible.*

The trial monitor will perform a closeout visit at the time when all CRFs have been retrieved and all queries have been answered.

18.5.2 *Electronic Case Report Forms*

Electronic Case Report Forms (eCRFs) will be provided for each participant. The trial monitor will review the forms at each site visit according to the monitoring plan.

Electronic Case Report Forms must be completed for all participants who sign Informed Consent, even if the participant fails to complete the trial. No section of the eCRFs is to be left blank without an appropriate explanation by the Investigator, since the lack of such explanation may necessitate discarding an otherwise usable observation. Instructions on how to fill in eCRF will be included in specific eCRF completion guidelines and Investigators will receive adequate training on them and on the electronic system (training will be required to get access to the system). Different accesses will be granted, depending on the site staff role in the trial.

If requested, access to eCRFs is to be granted to the appropriate regulatory agencies.

18.5.3 *Auditing/Inspecting*

The Investigator will make all pertinent records available, including source documentation, for inspection by regulatory authorities and for auditing by the Sponsor. This information will be considered as confidential. Audits/Inspections may occur any time from start to after conclusion of the trial. When an Investigator signs the protocol, they agree to allow regulatory authorities and Quince Therapeutics S.p.A auditors to inspect their trial records.

18.6 Archiving of Records

Copies of the protocol, participant identification codes, eCRFs, source data, ICFs and other documents pertaining to the trial conduct must be kept for the maximum period of time as required by the trial center. This time period must be at least 2 years after the last approval of the marketing application of the trial treatment in an ICH region and until there are no pending or contemplated marketing applications in an ICH region, or at least 2 years have elapsed since the formal discontinuation of clinical development of the trial treatment.

No trial document should be destroyed without prior written agreement between the Sponsor and the Investigator. Originals of all documentation and copies of outgoing correspondence concerning the trial will be stored and retained by the Sponsor in a safe area in the Trial Master File for the lifetime of the product. In particular, the final report sent by the Investigator to the IRB/IEC must be retained by the Sponsor, or the subsequent owner, for 5 years beyond the lifetime of the trial treatment.

18.7 Final Clinical Trial Report

The results of the trial will be reported in a Clinical Trial Report. This report will be prepared according to the Sponsor or delegated CRO SOPs and to the regulatory guidelines (ICH E3 Structure and Content of Clinical Trial Reports [Current Version]).

In compliance with the regulations, the final report will be communicated to the Member States where the trial was conducted/had investigational sites, within one year after end of the trial.

The final report will also be provided to the Investigators of the trial.

The summary of the results of the clinical trial will be accompanied by a summary written in a manner that is understandable to laypersons, according to the regulation (EU) No 536/2014 ("EU CTR").

18.8 Trial Documentation and Publication of Results

18.8.1 *Trial Documentation*

All unpublished documentation (including the protocol, CRFs and Investigator's Brochure) given to the Investigator is strictly confidential. All recipients must agree not to disclose the information herein contained to any person not connected with the trial without the prior written authorization of Quince Therapeutics S.p.A. The submission of these documents to the IRB/IEC is expressly permitted. The involved parties agree that the results of this trial will be used in their original form and/or in a global report for submission to governmental and regulatory authorities of any country.

All information communicated to the investigator(s) by Quince Therapeutics S.p.A is the exclusive property of Quince Therapeutics S.p.A. The PI will ensure this information shall be kept strictly confidential by him/her or any other person connected with the trial and shall not be disclosed to any third party without the prior written consent of Quince Therapeutics S.p.A.

18.8.2 *Publication of Results*

Any formal presentation or publication of the data from this trial will be considered as a joint publication by the Investigator(s) and Quince Therapeutics S.p.A. Authorship will be determined by a Publication Committee consisting of the lead investigator(s) from the trial, representatives from Quince Therapeutics S.p.A, and an external consultant with expertise in the field. For multi-center studies, it is mandatory that the first publication is based on data from all centers, analyzed as stipulated in the protocol by a statistician designated by Quince Therapeutics S.p.A. Investigators participating in this trial agree not to present or publish data gathered from a single center or sub-group of centers before the full initial publication, unless agreed to by all other investigators and Quince Therapeutics S.p.A. Authorship of any publication resulting from this trial will include members of each of the contributing centers and key contributors to the design and execution of the trial, including Quince Therapeutics S.p.A personnel.

Quince Therapeutics S.p.A will form a trial publication committee to coordinate and develop a publication policy and help in its implementation. The publication committee will be comprised of some PIs (to be determined by Quince Therapeutics S.p.A), based on their contributions to trial conduct and their status in the field, and key opinion leaders. The other members of the publication committee will include at least one representative of Quince Therapeutics S.p.A and an external consultant. Members of the publication committee cannot serve as first authors of more than one primary publication. The Publication Committee will decide on the content, journal, and sequence of publications /presentations.

The authorship of the first multicenter paper will comprise members of the publication committee, PIs in descending order of number of valid participants, and Quince Therapeutics S.p.A representatives who have made significant contributions to the design, conduct and analysis of the results.

Any publication, abstract, or paper of any information or material relating to or arising out of the present clinical trial shall be sent to Quince Therapeutics S.p.A for review at least 60 days before presentation at any congress, or publication of the final form(s) by any journal. Quince Therapeutics S.p.A will inform the Investigator of any changes or deletions necessary to preserve Quince Therapeutics S.p.A's confidential and proprietary technical information. All rights and interests worldwide in any inventions, know-how or other intellectual or industrial property rights, which arise during the course and/or as a result of the present clinical trial or which arise from the information or materials supplied under this Agreement, shall be assigned to, vest in and remain the property of Quince Therapeutics S.p.A.

18.9 Financial Agreement

A financial agreement (separate from the protocol) will be made and signed by Quince Therapeutics S.p.A or their designee and by a representative of the Institution where the trial will be conducted.

18.10 Termination of Trial

In the event that the Investigator is unable to continue the trial, another suitable sub-Investigator at the site will be designated to serve as the Investigator in the interim, until a new Investigator can be identified. This interim Investigator, if approved by the site IRB/IEC, will carry out the responsibilities of the Investigator. Documentation testifying to this will be submitted to the trial monitor within 10 days of the change. Within 6 months of the appointment of the interim Investigator, the new PI must be identified and approved by both Quince Therapeutics S.p.A and the IRB/IEC for the trial to continue at the site.

If the Sponsor and/or the Investigator should discover conditions arising during the trial that indicate it should be terminated, an appropriate schedule for termination will be instituted. If the Investigator terminates the trial, an explanatory letter will be provided to Quince Therapeutics S.p.A. Should the trial be discontinued due to a decision by Quince Therapeutics S.p.A, the Investigator will be reimbursed for reasonable expenses incurred and for the participants actually treated according to the trial protocol.

18.10.1 *Trial Discontinuation by the Sponsor*

The Sponsor may terminate the entire trial, or the trial at an individual site, at any time, for any of the following reasons:

- failure to enroll participants
- protocol violations or deviations
- inaccurate or incomplete data
- non-GCP compliance
- completion of enrolment
- administrative reasons

18.10.2 *Trial Discontinuation by the Investigator*

The Investigator may terminate their participation in the trial in consultation with the Sponsor due to the occurrence of significant AEs and/or adverse drug reactions endangering the health of participants, which make it ethically unacceptable to continue.

18.11 Insurance Policy

Quince Therapeutics S.p.A, or its designee, will provide insurance coverage for damages emerging from the trial and involving the participants treated with the test compound, provided that the Investigator(s) have adhered to the terms and provisions of the protocol. The principal Investigator will be supplied with all data concerning the insurance company and policy number for a maximum sum insurable.

18.12 Financial Disclosure

The PI and sub-Investigators will provide the Sponsor with adequate and accurate financial information (PD35) to ensure that the Sponsor can make complete and accurate financial certification of disclosure statements to concerned regulatory authorities. It is the duty of the Investigator to promptly update previous information provided to the Sponsor if there are salient changes that occur during the course of the trial, and for a period of one year following its completion (last participant last visit).

The trial will be performed under a US IND; therefore, all Investigators/sub-Investigators, contractors, etc., are expected to comply with the obligations as specified in the CFR (21 CFR part 54) by the US FDA, including requirements for full Financial Disclosure (Guidance for Clinical Investigators, Industry and FDA Staff: Financial Disclosure by Clinical Investigators, February 2013).

19 REFERENCES

Annese V, Latiano A, Rossi L, et al. Erythrocytes-mediated delivery of Dexamethasone in steroid-dependent IBD patients – A pilot uncontrolled study. Am J Gastroenterol 2005;100:370- 1375.

Boder E. Ataxia-telangiectasia: an overview. In: Gatti RA, Swift M, eds. Ataxia-telangiectasia: Genetics, Neuropathology, and Immunology of a Degenerative Disease of Childhood. Kroc Conference Series. Vol. 19. New York: Alan R. Liss Inc 1985:1-63.

Boehrs JK, He J, Halaby MJ, Yang DQ. Constitutive expression and cytoplasmic compartmentalization of ATM protein in differentiated human neuron-like SH-SY5Y cells. J Neurochem 2007;100(2):337-345.

Bossa F, Latiano A, Rossi L, et al. Erythrocyte-mediated delivery of dexamethasone in patients with mild-to-moderate ulcerative colitis, refractory to mesalamine: a randomized, controlled study. Am J Gastroenterol 2008;103:1-8.

Bossa F, Annese V, Valvano MR, et al. Erythrocyte-mediated delivery of Dexamethasone 21- phosphate in steroid-dependent Ulcerative Colitis: a randomized, double-blind, sham-controlled study. Inflamm Bowel Dis 2013;19:1872-79.

Broccoletti T, Del Giudice E, Amorosi S, et al. Steroid-induced improvement of neurological signs in ataxia-telangiectasia patients. Eur J Neurol 2008;15(3):223-8.

Broccoletti T, Del Giudice E, Cirillo E, et al. Efficacy of very-low-dose betamethasone on neurological symptoms in ataxia-telangiectasia. Eur J Neurol 2011;18(4):564-70. Epub 2010 Sep 14.

Buoni S, Zannolli R, Sorrentino L, Fois A. Betamethasone and improvement of neurological symptoms in ataxia telangiectasia. Arch Neurol 2006;63:1469-1482.

Busner J, Targum S, The Clinical Global Impressions Scale: Applying a Research Tool in Clinical Practice. Psychiatry, 2007.

Cano SJ, Hobart JC, Hart PE, et al. International Cooperative Ataxia Rating Scale (ICARS): appropriate for studies of Friedreich's ataxia? Mov Disord 2005;20(12):1585-91.

Castro M, Knafelz D, Rossi L, et al. Periodic treatment with autologous erythrocytes loaded with dexamethasone 21-phosphate for fistulizing pediatric Crohn's disease: Case report. J Pediatric Gastroenterol Nutr 2006;42:313-315.

Castro M, Rossi L, Papadatou B, et al. Long-term treatment with autologous red blood cells loaded with dexamethasone 21–phosphate in pediatric patients affected by steroid-dependent Crohn disease. J Pediatric Gastroenterol Nutr 2007;44:423-426.

Chessa L, Leuzzi V, Plebani A, et al. Intra-erythrocyte infusion of dexamethasone reduces neurological symptoms in Ataxia Telangiectasia patients: Results of a Phase 2 trial. Orphanet J Rare Dis 2014;9:5.

Chun HH, Gatti RA. Ataxia-telangiectasia, an evolving phenotype. DNA Repair 2004;3:1187- 1196.

Clinical Trials Facilitation and Coordination Group (CTFG). Recommendations related to contraception and pregnancy testing in clinical trials; Version 1.1, 21 Sept 2020.

Crawford TO, Skolasky RL, Fernandez R, et al. Survival probability in ataxia telangiectasia. Arch Dis Child 2006;91(7):610-11.

Crinelli R, Antonelli A, Bianchi M, Gentilini L, Scaramucci S, Magnani M. Selective inhibition of NF-kB activation and TNF-α production in macrophages by red blood cell-mediated delivery of dexamethasone. Blood Cells Mol Dis 2000;26(3):211-222.

D'Ascenzo M, Antonelli A, Chiarantini L, Mancini U, Magnani M. Red blood cells as a glucocorticoids delivery system. In: Erythrocytes as drug carriers in medicine. Sprandel U, Way JL (eds), Plenum Press, New York 1997,81-88.

Dumont LJ, AuBuchon JP. Evaluation of proposed FDA criteria for the evaluation of radiolabelled red cell recovery trials. Transfusion 2008;48:1053-60

EryDex System Investigator's Brochure, current version

Gordon CM et al., 2013 Pediatric Position Development Conference: Executive Summary and Reflections. Journal of Clinical Densitometry: Assessment and Management of Muscoloskeletal Health, 2014; 17, (2): 219-224.

Guy W. Clinical Global Impressions. 1976. National Institute of Mental Health. ECDEU Assessment Manual for Psychotherapy.

Herdman M, Gudex C, Lloyd A, et al. Development and preliminary testing of the new five-level version of the EQ-5D (EQ-5D-5L). Qual Life Res 2011;20(10):1727-36.

Hoche F, Seidel K, Theis M, et al. Neurodegeneration in Ataxia Telangiectasia. What is New? What is Evident? Neuropediatrics 2012;43(3):119-29. Epub 2012May21.

Howie SR. Blood sample volumes in child health research: review of safe limits. Bull World Health Organ 2011;89:46-53.

Jackson TJ, Chow G, Suri M, Byrd P, Taylor MR, Whitehouse WP. Longitudinal analysis of the neurological features of ataxia-telangiectasia. Dev Med Child Neurol. 2016 Jul;58(7):690-7. doi: 10.1111/dmcn.13052. Epub 2016 Feb 19. PMID: 26896183.

Janssen M, Pickard AS, Golicki D, et al. Measurement of properties of the EQ-5D-5L compared to the EQ-5D-3L across eight patient groups: a multi-country study. Qual Life Res 2013;22(7):1717-27.

Lee Y, McKinnon PJ. ATM dependent apoptosis in the nervous system. Apoptosis 2000;5(6):523-529.

Lucidi V, Tozzi AE, Bella S, Turchetta A. A pilot trial on safety and efficacy of erythrocyte- mediated steroid treatment in CF patients. BMC Pediatrics 2006, 6:17.

Lynch DR, Perlman SL, Meier T. A phase 3, double-blind, placebo-controlled trial of idebenone in Friedreich ataxia. Arch Neurol 2010;67(8):941-7.

Magnani M, Rossi L, D'Ascenzo M, et al. Erythrocyte engineering for drug delivery and targeting. Biotechnol Appl Biochem 1998;28:1-6.

Mambrini G, Mandolini M, Rossi L, Pierigè F, Capogrossi G, Salvati P, Serafini S, Benatti L, Magnani M. Ex vivo encapsulation of dexamethasone sodium phosphate into human autologous erythrocytes using fully automated biomedical equipment. Int J Pharm. 2017 Jan 30;517(1-2):175-184. doi: 10.1016/j.ijpharm.2016.12.011. Epub 2016 Dec 7. PMID: 27939571.

Marshall WA, Tanner JM (June 1969). Variations in pattern of pubertal changes in girls. Arch. Dis. Child. 44 (235): 291–303.

Marshall WA, Tanner JM (February 1970). Variations in the pattern of pubertal changes in boys. Arch. Dis. Child. 45 (239): 13–23.

Meier T, Perlman SL, Rummey C, et al. Assessment of neurological efficacy of idebenone in pediatric patients with Friedreich's ataxia: data from a 6-month controlled study followed by a 12- month open-label extension study. J Neurol 2012;259(2):284-91.

Menotta M, Biagiotti S, Bianchi M, et al. Dexamethasone partially rescues ataxia telangiectasia- mutated (ATM) deficiency in ataxia telangiectasia by promoting a shortened protein variant retaining kinase activity. J Biol Chem 2012;287(49):41352-63.

Metz G, Coppard N, Cooper JM, et al. Rating disease progression of Friedreich's ataxia by the International Cooperative Ataxia Rating Scale: analysis of a 603-patient database. Brain 2013;136(Pt 1):259-68.

Mitchell CD, Richards SM, Kinsey SE, et al.; Medical Research Council Childhood Leukaemia Working Party. Benefit of dexamethasone compared with prednisolone for childhood acute lymphoblastic leukaemia: results of the UK Medical Research Council ALL97 randomized trial. Br J Haematol 2005;129:734-745.

Natale, V.A.I., Cole, T.J., Rothblum-Oviatt, C. et al. Growth in ataxia telangiectasia. Orphanet J Rare Dis 16, 123 (2021). https://doi.org/10.1186/s13023-021-01716-5.

Nissenkorn A, Hassin-Baer S, Lerman SF, et al. Movement disorder in ataxia-telangiectasia: treatment with amantadine sulfate. J Child Neurol 2013;28(2):155-60.

Nissenkorn A, et al., Development of global rating instruments for pediatric patients with ataxia telangiectasia, European Journal of Paediatric Neurology (2015), http://dx.doi.org/10.1016/j.ejpn.2015.09.002

Orphanet Report Series - Prevalence of rare diseases: Bibliographic data - May 2014 - Number 1 http://www.orpha.net/orphacom/cahiers/docs/GB/Prevalence_of_rare_diseases_by_ alphabetical_list.pdf

Perlman S, Becker-Catania S, Gatti RA. Ataxia-telangiectasia: diagnosis and treatment. Semin Pediatr Neurol 2003;10:173-182.

Petley E, Yule A, Alexander S, et. al. The natural history of ataxia-telangiectasia(A-T): A systematic review. PLOS ONE 2022 March 15. doi:10.1371/journal.pone.0264177

Posner K, Oquendo MA, Gould M, et al. Columbia Classification Algorithm of Suicide Assessment (C-CASA): Classification of suicidal events in the FDA's pediatric suicidal risk analysis of antidepressants. Am J Psychiatry 2007; 164:1035-1043.

Posner K, Brown GK, Stanley B, et al. The Columbia-Suicide Severity Rating Scale: Initial Validity and Internal Consistency Findings from Three Multisite Studies with Adolescents and Adults. Am J Psychiatry 2011; 168:1266-1277.

Poupard M. Ataxia-telangiectasia. In: Euro-Ataxia: European Federation of Hereditary Ataxia [newsletter 24]. 2003:7-9.

Rabin R, de Charro F. EQ-5D: A measure of health status from the EuroQol Group. Annals of Medicine 2001;33:337–343.

Rossi L, Serafini S, Cenerini L, et al. Erythrocyte-mediated delivery of dexamethasone in patients with chronic obstructive pulmonary disease. Biotechnol Appl Biochem 2001;33:85-89.

Rossi L, Castro M, D'Orio F, et al. Low doses of dexamethasone constantly delivered by autologous erythrocytes slow the progression of lung disease in cystic fibrosis patients. Journal of Blood Cells Mol Dis 2004;33(1):57-63.

Rothblum-Oviatt C, Wright J, Lefton-Greif MA, McGrath-Morrow SA, Crawford TO, Lederman HM. Ataxia telangiectasia: a review. Orphanet J Rare Dis. 2016 Nov 25;11(1):159.

Russo I, Cosentino C, Del Giudice E, et al. In ataxia-teleangectasia betamethasone response is inversely correlated to cerebellar atrophy and directly to antioxidative capacity Eur J Neurol 2009;16:755-759.

Schmahmann JD, Gardner R, MacMore J, Vangel mg. Development of a Brief Ataxia Rating Scale (BARS) based on a modified form of the ICARS. Mov Disord 2009;24(12):1820-8.

Schmitz-Hubsch T, du Montcel ST, Baliko L, et al. Reliability and validity of the International Cooperative Ataxia Rating Scale: a study in 156 spinocerebellar ataxia patients. Mov Disord 2006a;21(5):699-704.

Schmitz-Hubsch T, du Montcel ST, Baliko L, et al. Scale for the assessment and rating of ataxia: development of a new clinical scale. Neurology 2006b;66(11):1717-20.

Schoch B, Regel JP, Frings, et al. Reliability and validity of ICARS in focal cerebellar lesions. Mov Disord 2007;22(15):2162-9.

Trouillas P, Takayanagi T, Hallet M, et al. International Cooperative Ataxia Rating Scale for pharmacological assessment of the cerebellar syndrome. The Ataxia Neuropharmacology Committee of the World Federation of Neurology. J Neurol Sci 1997;145(2):205-11.

Weyer A, et al. Reliability and validity of the scale for the assessment and rating of ataxia: a study in 64 ataxia patients. Mov Disord 2007;22(11):1633-7.

Zannolli R, et al. A randomized trial of oral betamethasone to reduce ataxia symptoms in ataxia telangiectasia. Mov Disord 2012;27(10):1312-6.

Zocchi E, Guida L, Polvani C, et al. Human and Murine Erythrocytes as Bioreactors Releasing the Antineoplastic Drug 5-fluoro-2'-deoxyuridine. In Resealed Erythrocytes as carriers and bioreactors. Advances in the Biosciences (Volume 81), Pergamon Press, R. Green, J.R. DeLoach (1991).

APPENDICES

Appendix 1: Investigator Statement

Investigator Statement

Investigational Medicinal Product:	EryDex System
Protocol Number:	IEDAT-04-2022
Title:	A Multi-center, Randomized, Double-blind, Placebo-controlled Trial to Evaluate the Neurological Effects of EryDex on subjects with Ataxia-Telangiectasia
Principal Investigator:	
Trial Center:	

COMMITMENTS

By signing this document, I agree to conduct the trial as outlined in the protocol and in accordance with (as applicable) the Declaration of Helsinki, the ICH GCP Guideline as well as all applicable government regulations.

I declare:

1. I am well qualified by scientific training and experience to conduct investigational studies in the clinical area of the proposed trial and I am affiliated with a recognized medical school or with an independent institution recognized for its excellence.
2. I have received and understood the information about pharmacology, toxicology and possible risks and side effects of the investigational compound (e.g., as described in the Investigator's Brochure).
3. I shall provide information to all staff members involved in the trial about their obligations as described in this document.
4. I shall submit the protocol, Informed Consent form/participant information sheet and other required documentation to the IRB/IEC for review and approval.
5. I shall make no changes to the protocol without formal amendment (prepared in agreement with the Sponsor), except when necessary to protect the safety, the rights or welfare of participants. In this last case I will inform the Sponsor of the change.
6. I shall require Informed Consent from each participant prior to enrolment into the trial. The Informed Consent shall be documented by use of a written consent form approved by the Sponsor and the IRB/IEC.
7. I shall use the investigational compound only in compliance with the trial protocol and I shall be responsible for the security and accountability of clinical trial supplies.
8. I shall notify Quince Therapeutics S.p.A immediately or no later than 24 hours by telephone serious or unexpected AEs and submit written reports of AEs, as outlined in the protocol, to Quince Therapeutics S.p.A, the IRB/IEC and to Regulatory Authorities (when appropriate).
9. I shall complete the Sponsor's CRF in a timely and legible manner.

10. I shall maintain accurate source records (hospital or other institutional records), which will support the data entered into CRF and I shall maintain these as specified in the protocol.
11. I shall allow monitoring visits by Quince Therapeutics S.p.A's representatives at predetermined frequency.
12. I shall allow the authorized Sponsor representative and any Regulatory Authorities to inspect the facilities and pertinent records at reasonable times and in a manner which ensure participant confidentiality.
13. I shall maintain confidentiality about all information concerning the investigational compound, such as patent applications, formulas, manufacturing process, basic scientific data and formulation information supplied by the Sponsor and not previously published and I shall not disclose this information to a third party without the written consent of the Sponsor.
14. I shall permit the information developed in the clinical trial to be used by the Sponsor in connection with the development of the compound and may be disclosed to the IRB/IEC and Regulatory Authorities.

Following completion of the trial, the data may be considered for reporting at a scientific meeting and/or for publication in a scientific journal. A copy of the manuscript or abstract will be provided to the Sponsor for review before submission to a scientific journal for publication and/or a scientific meeting selection committee for oral or poster presentation. Subgroup or individual Investigator publications must not interfere or compromise publication of the multi-center results of this clinical trial.

_____ _____

Investigator Signature **Date**

_____

Investigator (Printed Name)

Appendix 2: Description of the EryDex System

The EDS is used for the ex vivo encapsulation of the drug DSP into autologous erythrocytes before re-infusion of the drug-encapsulated erythrocytes into a participant. [redacted]

EDS Components:

The EDS comprises the following:

- RCL Device (CE Marked)
- EryKit_01 (Single-Use Device CE Marked)
- Syringe Kit (Single-Use Device CE Marked)
- Process Solutions :
  - Hypotonic Solution 1 (CE Marked)
  - Hypotonic Solution 2 (CE Marked)
  - PIGPA Hypertonic Solution (CE Marked)
- Drug: DSP Solution

The components are intended to be marketed for use solely with the EDS. Some components, such as the EryKit_01, Syringe Kit, Process Solutions and Drug are single-use; the RCL device is reusable.

Red Cell Loader Device

[redacted]

EryKit_01 Device

[redacted]

Syringe Kit Device

[redacted]

Process Solutions

[redacted]

[redacted]

Dexamethasone Sodium Phosphate Solution 25 mg/mL

[redacted]

[redacted]

Additional materials

Additionally, the following standard, widely available materials (not provided as a part of the EDS) are required for the proper operation of the EDS:

[redacted]

EryDex System Process (or Procedure)

[redacted]

[redacted]

[redacted]

[redacted]

[redacted]	[redacted]
[redacted]	[redacted]
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[redacted]

Appendix 3: Method for Measuring Dexamethasone Sodium Phosphate in Samples Taken from Satellite Sample Bag or any other EryDex Sampling Point

Appendix 4: Procedure for Collection of Blood Samples for Laboratory Measurement of Free Plasma Hemoglobin (Hemolysis Panel)

[redacted]

Below are reported some potential causes of in vitro hemolysis that must be avoided. In case one or more of these problems occurs, sampling must be repeated:

[redacted]

References:

1. Dugan L, Leech L, Speroni KG, Corriher J. Factors affecting hemolysis rates in blood samples drawn from newly placed IV sites in the emergency department. J Emerg Nurs. 2005 Aug;31(4):338-45.
2. Halm MA, Gleaves M. Obtaining blood samples from peripheral IV catheters: best practice? Am J Crit Care. 2009 Sep;18(5):474-8.
3. Lippi, et al. Hemolyzed specimens: a major challenge for emergency departments and clinical laboratories. Critical Reviews in Clinical Laboratory Sciences, 2011;48(3):143–153.

Appendix 5: Sterility Test of EryDex Using a Culture-based Method – Sampling Procedure

Results Classification:

- TRUE POSITIVE: Initial positive, confirmed by growth of same bacteria on repeat culture of original specimen.
- FALSE POSITIVE: Initial positive, no growth on repeat culture of original specimen.
  INDETERMINATE: Initial positive, with no confirmatory test performed or confirmatory test performed but cannot be interpreted.

At the trial's start, investigational sites should collect and provide to the Sponsor/CRO the local procedure that will be followed for the conduct and interpretation of the culture-based method sterility test, including any available quality certificate.

Appendix 6: Action Plan in Case of Positive Post-Release Sterility Test Results

MANAGEMENT OF THE PARTICIPANT

- The laboratory will immediately inform the PI (or the designee) by phone and e-mail of the result of the culture-based test; the PI will immediately send this information to the Sponsor/CRO. The Sponsor will immediately inform the Chairman of the iDSMB.
- The investigator/staff member will immediately contact the transfused participant and/or their caregiver, to determine if there are any signs of bloodborne infection (bacteremia or sepsis), e.g., fever, back or flank pain, skin flushing, pallor, blood in urine, tachycardia, chills, fainting or dizziness, etc. The participant/caregiver will be asked to contact their healthcare professional and inform them of the finding. The healthcare professional must be informed that the participant is receiving monthly IV infusions that may contain DSP.
    - If the participant is asymptomatic, the local healthcare professional should evaluate the participant and take blood samples for a series of tests that include a rapid microbial test, samples for culture*/sensitivity, ESR and a CBC. If the participant remains asymptomatic for the next 48 hours, and the results of the clinical evaluation and CBC do not indicate signs of an active infection, the PI should determine the need for further evaluation until the next visit. All the documentation relevant to the investigations (lab reports, medical notes) should be made available at the investigational site.
    - If the participant is symptomatic, the caregiver will be asked to take the participant to the nearest hospital immediately or return to the investigational site (if feasible), for further management and evaluation of the participant. The hospital laboratory will perform all the necessary tests to confirm/identify the infective agent. These tests will include a rapid microbial test, sampling for culture*/sensitivity, ESR and a CBC. The results of this culture will be compared to determine if the identified infectious agent is the same as the one from the culture of the EryDex (Note: Investigational sites should collect and provide to the Sponsor/CRO the local procedure that will be followed for the conduct and interpretation of the culture, including any available quality certificate). The participant should be treated with the most appropriate anti-infective agent based on the symptoms and sensitivity results from the blood culture. The PI will evaluate the participant 2 weeks after resolution of all symptoms, and if blood tests are normal, will determine if the participant can continue the investigational treatment. All the documentation relevant to the investigations (lab reports, medical notes) should be made available at the investigational site and provided to the Sponsor/CRO (after being adequately de-identified) as needed, to perform the required investigations and comply with any reporting requirements.

(*) the pre-infusion blood culture performed at the subsequent Protocol visit is not considered a valid alternative to such measures, unless the subsequent visit is scheduled a few days after receiving the alert from the local laboratory of the positive EryDex sample culture base sterility test relevant to the previous treatment visit.

ACTIONS TO BE PERFORMED AT THE INVESTIGATIONAL SITE

Whenever the result from the EryDex culture indicates a contamination, a series of actions will be performed at the concerned site:

- The hospital Laboratory will perform the investigation to identify the EryDex contaminant.
- The EryDex process records relevant to the infusion with the positive EryDex test will be reviewed to determine if there were any discrepancies, mishaps, missteps, or differences in the procedures followed for the participant whose culture provided a positive result.
- The site will be asked to complete an EryDex process using all EryDex components including IMP; the EryDex process will be performed with sterile saline solution instead of participant blood. The resulting sample will be submitted to the hospital Laboratory for culture/sensitivity analyses.
- The site will be required to review the procedures to be followed to ensure sterile conditions are maintained throughout the EryDex process. In addition, precautions will be reviewed to ensure the sampling and testing of EryDex are done under sterile conditions and applying aseptic procedures.
- Site staff will report positive results as AEs or SAEs, as appropriate, specifying the sample that tested positive (e.g., EryDex).

ACTIONS BY THE SPONSOR

The Sponsor, upon receiving the information that the results of the culture tested positive for microbial growth, will undertake a series of actions:

- The Sponsor will inform the Data Safety Monitoring Board, the FDA, and other relevant Health authorities, according to the local reporting procedures and timelines.
- Investigate the cause and take corrective actions. The Sponsor will review records relating to the shipping of the investigational treatment and products to the site and their storage at the site. Training records will be examined to ensure that operators received adequate training and will also review the EryDex process records. Sponsor representatives may attend new infusions to ensure that all procedures are followed.
- Positive results reported as AEs or SAEs will be analyzed and included in the IB, DSUR and any other relevant regulatory/safety documents.

Appendix 7: Signs and Symptoms of Adrenal Insufficiency

Screening for adrenal insufficiency will be performed in all participants, via early morning (before 8:00 AM) plasma cortisol testing, at the following times: (1) during the screening period (prior to randomization), (2) when participants are symptomatic, and (3) when participants are at most risk for adrenal insufficiency and adrenal crisis, such as following discontinuation or interruptions of dexamethasone dosing (i.e., loading failures and following discontinuation of the trial treatment).

The following signs and symptoms will be assessed to screen for adrenal insufficiency:

- Extreme fatigue
- Weight loss and decreased appetite
- Darkening of skin (hyperpigmentation)
- Low BP, even fainting
- Salt craving
- Hyponatremia
- Hyperkalemia
- Low blood sugar (hypoglycemia)
- Nausea, diarrhea, or vomiting
- Abdominal pain
- Muscle or joint pains
- Irritability
- Depression
- Confusion
- Body hair loss or sexual dysfunction in women

Clinical judgement should be used by the investigator to determine if any of the above signs and symptoms observed are indicative of adrenal insufficiency related to the trial treatment or are symptoms of the underlying neurological disorder.

If the basal (8:00 AM) cortisol level is measured and is within the reference normal range, the participant can be enrolled in the trial or can continue dosing with EryDex. If the 8:00 AM cortisol level is below 3-5 µg/dL (depending on assay) regardless of symptoms, or the participant exhibits signs or symptoms of adrenal insufficiency and has a cortisol level <10 µg/dL, the participant will receive a high-dose ACTH stimulation test as soon as possible. If the ACTH stimulation test is normal, the participant can be enrolled, or can continue dosing with EryDex, after the effects of the ACTH have dissipated (e.g., 30 days after ACTH is administered, resulting in a prolongation of the Screening period). If the participant fails the ACTH stimulation test, they will be excluded or discontinued from the trial (not applicable for Visit 9 testing) and referred to an endocrinologist (pediatric endocrinologist, depending on the participant's age) with a recommendation to prescribe stress dose steroids.