INFORMATION SHEET FOR PARENT(S) OR LEGAL GUARDIAN TO INCLUDE A MINOR IN A CLINICAL TRIAL

Official title of the trial *Newborn screening for the early diagnosis of Metachromatic Leukodystrophy - MLD*
Official title of the trial in more patient-friendly terms: Development of a screening test for all newborns in Lombardy to identify patients suffering from Metachromatic Leukodystrophy (MLD), a rare disease that can lead to the patient's death. Early treatment, however, can save the patient's life.
Facility-context in which the trial will take place: Newborn Screening, Functional Genomics and Rare Diseases Unit
Coordinating Centre *(if different from the facility hosting the trial)* **and trial coordinator** Coordinating centre: Newborn Screening, Functional Genomics and Rare Diseases Unit Trial coordinator: Dr. Cristina Cereda
Registry in which the trial has been or will be registered (if applicable) and identification code if available Identification code _____ Registry_____
Main Investigator *(indicate the local trial Investigator)* Name _____ Affiliation _____
Sponsor/Funding body/Foundation Fondazione Buzzi on the basis of a Telethon grant
Ethics Committee Lombardy Territorial Ethics Committee 1

This document consists of the following sections:

A. INTRODUCTION
B. INFORMATION SECTION. SUMMARY OF THE TRIAL: KEY INFORMATION
C. INFORMATION SECTION. ADDITIONAL INSIGHTS
D. EXPRESSION OF CONSENT SECTION

ATTACHMENTS

ADDITIONAL DOCUMENTS *Dear Sir/Madam, the information contained in the following information sheet is very detailed. We are asking you to agree on taking part in the trial ONLY after you have read this information sheet carefully and have had a COMPREHENSIVE TALK with a member of the trial team, who will have to dedicate the NECESSARY TIME for you to understand in full what we are proposing.*

A. INTRODUCTION

Dear Madam/Sir (Guardian),

We are proposing the participation of your child/the minor in the clinical trial, which is described below.

It is your right to be informed about the purpose and characteristics of the trial so that you can make an informed and free decision on whether to authorise participation.

The purpose of this document is to inform you about the nature of the trial, its purpose and what participation in it will entail, including your rights and responsibilities.

Please read the following carefully. The researchers involved in this project, indicated at the beginning of this document, are available to answer your questions. No question that comes to your mind is trivial: do not be afraid to ask it!

In addition to us, you can discuss the proposal in this document with your family doctor/paediatrician, your family and any other people you trust. Take as much time as necessary to decide. You can take home an unsigned copy of this document to reflect on it or to discuss it with others before making a decision.

If you decide that your child/the minor will not participate in the trial, your child/the minor will still receive the best possible care for patients with his/her condition/disease.

Your refusal will in no way be interpreted as a lack of trust.

In order to make this document easier to understand, the Trial Centre provides versions of it in the most widely spoken languages.

If you are unable to sign the informed consent, consent may be given and recorded by alternative means, e.g. audio or video recordings in the presence of at least one impartial witness.

Once you have read this form, received answers to any questions you may have and decided to authorise your child's/the minor's participation in the trial, you will be asked to sign a consent form, of which you will receive a paper copy.

The Main Investigator

B. INFORMATION SECTION.

GENERAL SUMMARY OF THE TRIAL: KEY INFORMATION

Metachromatic Leukodystrophy (MLD) is a lethal hereditary disease caused by lysosomal storage disorders linked to mutations in the ARSA gene, which codes for the enzyme arylsulfatase A. Alteration of this enzyme results in the accumulation of sulfatides in lysosomes, small organelles within the cells of the central nervous system, peripheral nervous system and other tissues. This accumulation causes the loss of the myelin sheath that surrounds nerve cells, leading to the loss of motor and cognitive functions, and even to death, especially when the disease has an early onset. The incidence of this disease is 1.1 cases per 100,000 births in Europe, which allows it to be defined as a rare disease. There are different forms according to the time when motor and cognitive symptoms show up: the infantile form (within 30 months of life), the early juvenile form (between 2½ and 6 years of age), the late juvenile form (between 6 years and 16 years of age) and the adult form. The first two forms are the most severe. However, there are therapies that can delay the onset and mitigate the progression of this disease. Such therapies include haematopoietic stem cell (HSC) transplantation, gene therapy by autologous transplantation (GT) – which is specific to the juvenile and adult forms of MLD and which aims at transplanting autologous CD34+ cells translated with a lentivirus in order to obtain the overexpression of the ARSA (arylsulfatase A) enzyme involved in pathogenic processes – and OTL-200 gene therapy specific for children and newborns. All of them are already available and authorised by the European Commission. Therefore, since early diagnosis is crucial, the Regional Reference Laboratory for Newborn Screening has set the primary goal of the present study: developing the analytical method required to diagnose MLD in the context of newborn screening,

starting from the blood sample taken on a card for mandatory newborn screening. This first goal is achieved by using a highly sensitive and specific method to assay the tandem mass spectrometry of accumulation products, i.e. sulfatides. If the first analysis is positive, it is repeated and the ARSA enzyme is assayed too. A positive result in the second analysis will require a second control card. In this case, the birth centre will contact you for the collection. Positivity to the second-level test will result in the patient being referred to the clinical reference centre, i.e. San Raffaele Hospital in Milan. Here, the patient can complete the diagnostic procedure by determining the ARSA activity on leucocytes and sulfatides in 24-hour urine and by performing a genetic sequencing test to detect two pathogenic mutations of the ARSA gene, subject to the provision of the informed consent for genetic testing by San Raffaele Hospital. The patient can also start treatment following the most suitable therapeutic strategies for the specific case.

This section aims to briefly present the key aspects of the trial we are proposing. The sections below will provide more details for you to give a fully informed consent to your child's/the minor's participation in the trial, or not.

- Why are you asking us to give consent for participation in this trial?

We are asking you to give your consent to participate in a clinical trial financed by Fondazione Buzzi on the basis of a Telethon grant because your child/the minor may be suffering from Metachromatic Leukodystrophy and the treatments available to date are life-saving, if administered in the early stages of life.

Your child/the minor has been included among those patients who are asked to take part in this trial because he/she shows some clinical features that will be described in more detail in section C.

- What are the goals of the trial? How many centres and patients will be part of it?

The trial is being conducted in order to answer the question 'Are the screened newborns positive for sulfatides and, therefore, are they potentially suffering from Metachromatic Leukodystrophy?'

INDICATE THE SECONDARY GOAL(S)

The trial will be performed at the Regional Reference Laboratory for Newborn Screening, which is part of the Newborn Screening, Functional Genomics and Rare Diseases Unit, and 100,000 patients will be included.

The secondary goals are to develop the test performed as a screening test: in the future, it may be included in the newborn screening programme that already exists in Lombardy for other rare diseases.

In addition, other family members of test-positive newborns will be screened for a possible diagnosis as well, since these diseases are genetically based.

Lastly, the study will make it possible to acquire epidemiological data, thus making it possible to estimate the incidence of Metachromatic Leukodystrophy in Lombardy.

- What is the routine treatment approach for treating our child's/the minor's disease?

Infants whose sulfatides test positive - being the subject of analysis in this study – will be referred to the Clinical Reference Centre, which will propose confirmatory diagnostic tests and therapeutic and monitoring strategies for the patient in the event of a positive result.

- Can I/we freely decide whether to participate or not?

You will be free to choose whether to agree to participate in the trial or not. Even after agreeing, you will be able to change your mind at any time.

- What choices do I/we have if I/we decide not to give consent for our child/the minor to participate in the trial?

If you decide not to allow your child/the minor to take part in the trial, they will, however, follow the newborn screening procedure according to the regulations in force in the Lombardy Region.

In addition, they will be able to participate in another trial that may be in progress.

- What happens if I/we decide to authorise participation in the trial?

If the patient decides to participate in the trial, they will not have to take any more samples than those already planned for the newborn screening programme. In fact, the biological material used for the sulfatides test is the residual material from the card used for newborn screening. As with all newborn screening tests, the patient will only be contacted in case of a positive result and referred to the Clinical Reference Centre at the IRCCS San Raffaele Hospital San Donato Group in Milan.

The full schedule of planned trial tests is given in the next section 'What examinations, tests and procedures are planned in the trial?'

- What are the risks and benefits if I/we decide to authorise participation in the trial?

Both risks and benefits may arise from participation in the trial. It is important to assess them carefully before making a decision. In reality, there are no risks in this trial, as the patient is not required to collect any further blood samples in addition to that already taken as part of the newborn screening programme. The benefit lies in having the opportunity of an early diagnosis for a potentially fatal disease, but for which an effective and life-saving therapy is available if administered early in life. The benefit is also for all the family members of the positive patient, who, alerted by the positive outcome of the newborn, have the opportunity to undergo diagnostic tests to investigate the presence (or absence) of the disease.

- Expected benefits

Benefits for the patient: *The benefits lie in having the opportunity for the early diagnosis of a potentially fatal disease, but for which an effective and life-saving therapy is available if administered early in life. The benefit is also for all family members of the positive patient, who, alerted by the positive outcome of the newborn, have the opportunity to undergo diagnostic tests to investigate the presence (or absence) of the disease.*

Benefits for other patients: *By joining the trial, your child/the minor will contribute to developing a new diagnostic test for their disease. In the future, other patients with their disease may benefit from it. In addition, the family members of the positive patient will have the opportunity to be diagnosed with the same disease.*

Potential risks

No potential risks are associated with participation in this study.

- Is consent final? Can I/we decide to withdraw our child/the minor from the clinical trial (voluntary withdrawal)?

You can decide to withdraw your child/the minor from the trial at any time and for any reason, without need to motivate your decision.

If you decide not to have them participate any more, please let one of the investigating doctors/biologists know as soon as possible.

The doctor/biologist will keep you informed of any changes in the trial that may influence your willingness to continue participation.

- Are there any reasons why the trial could be terminated other than by my/our will (early termination)?

Yes, the investigating doctor/biologist may decide to terminate your child's/the minor's participation in the trial if:

- *His/ her health condition changes and participation in the trial is potentially harmful*
- *New information becomes available and the trial is no longer in their best interest*
- *Your child/the minor does not comply with the agreed rules for participation in the trial*

- *The trial is terminated by the competent authorities or by the sponsor.*

IN CASE CONSENT IS WITHDRAWN AND/OR THE TRIAL SUSPENDED, THE PATIENT WILL STILL BE ABLE TO COMPLETE HIS/HER NEWBORN SCREENING PROGRAMME ACCORDING TO THE REGULATIONS IN FORCE.

C. INFORMATION SECTION. ADDITIONAL INSIGHTS

1. What is the purpose of the trial?

The aim of this study is to develop a method, which has so far only been tested in Tuscany at the Meyer Hospital by Prof. La Marca's group, to perform a first-level screening test for Metachromatic Leukodystrophy. In order to develop a method, it is necessary to carry out a number of tests so that it is possible to define normal ranges for threshold values above which it is possible to consider the patient positive to the performed test. In this specific case, the test that would make it possible to establish the presence or absence of the pathology under study is the determination of sulfatides on the drop of blood taken for newborn screening on a card. Positivity for the four sulfatides tested is not sufficient for a final diagnosis. Diagnostic confirmation tests are needed. Currently these tests can be performed by the Clinical Reference Centre at San Raffaele Hospital, which is the second centre involved in this study.

Which patient groups are being compared? What is the intervention being tested?

- *The present study can be summarised in two phases:*

FIRST PHASE

Newborns in the Lombardy region, for whom their parents/legal guardians have signed the informed consent, undergo the test at the regional reference laboratory for newborn screening at the Newborn Screening, Functional Genomics and Rare Diseases Unit of the 'V. Buzzi' Children's Hospital in Milan - ASST Fatebenefratelli Sacco.

SECOND PHASE

In the event of a positive result, the patient is referred to the clinical reference centre of San Raffaele Hospital in Milan, which is the second centre in this study and which is responsible for confirming the positive result obtained from the screening. They are doing so by measuring the ARSA activity in the leukocytes and sulfatides positivity in 24-hour urine, and by searching for ARSA gene mutations. Once a final diagnosis has been made, the clinical reference centre takes charge of the patient for treatment and monitoring.

- *Here follows a list of the criteria according to which a patient is included in or excluded from the study:*

INCLUSION CRITERIA

Newborns in the Lombardy region who undergo newborn screening, for whom their parents/legal guardians have signed the relevant informed consent.

EXCLUSION CRITERIA

Newborns in the Lombardy region who undergo newborn screening, for whom the parents/legal guardians have not signed the relevant informed consent.

- *Re-examination is performed at 48-72 hours of life, when the material for newborn screening is collected.*

  The blood drop is analysed by means of a tandem mass spectrometry using a ULPC-MS/MS system, ABI SCIEX API 4000 Perkin Elmer type, using a Kinetic C8 Column 2.6 µm 50x2.1 mm chromatography column. The chromatographic run and subsequent identification by tandem mass spectrometry allows

detecting four sulfatides (C16, C16-OH, C16:1 and C16:1-OH), which, in the case of disease, accumulate in lysosomes, i.e. small organelles within the cells in the central and peripheral nervous system and in other tissues.

- *Currently, an effective gene therapy exists for Metachromatic Leukodystrophy, which was approved by the European Union at the end of 2020: it can save the lives of patients for whom the accumulation of these toxic substances (sulfatides) would inevitably lead to death. However, there is no early diagnosis system in place across the entire population, and, being a rare disease, it would lend itself to screening in the newborn phase. This diagnostic deficiency convinced the Regional Reference Laboratory for Newborn Screening to embark on a pilot project to develop an analytical method to select newborns for this disease.*
- *In the event of a positive result in the sulfatides screening, the newborn will be contacted by its birth centre, as is already the case for mandatory neonatal screening, and will be referred to the Clinical Reference Centre for Metachromatic Leukodystrophy at San Raffaele Hospital in Milan.*
- *The study is defined as multicentre, as the two centres of the Newborn Screening, Functional Genomics and Rare Diseases Unit of the Hospital are in charge of performing the first-level newborn screening test while the Clinical Reference Centre for Metachromatic Leukodystrophy of San Raffaele Hospital is in charge of the diagnostic confirmation and administration of the therapy.*

Overview of the clinical trial

See scheme in Annex 1 to this form.

2. What examinations, tests and procedures are included if we authorise participation in the trial?

Newborns taking part in this study will not have to take further blood samples, in addition to that already taken at 48-72 hours of life for mandatory newborn screening according to the regulations in force in the Lombardy Region. In fact, the first-level screening test for MLD will be performed on the residual material resulting from this collection. Patients for whom the result of this test is negative will not undergo any further visits and/or examinations and the negative result will not be communicated, unless requested at their birth centre. Patients for whom the result of this test is positive will be referred to the clinical reference centre for MLD at the Paediatric Immunohematology Unit of the IRCCS San Raffaele Hospital San Donato Group in Milan. Here, the parents/legal guardian will be offered with one or more confirmation tests, including a genetic test, for which the reference clinical centre will give you specific consent forms to sign. In the event of a positive confirmation test result, the reference clinical centre will propose a gene therapy, which you may or may not choose to resort to, subject to adequate information and signing of the relevant informed consent forms. For any details on the frequency and duration of the visits, the planned examinations and therapies and the entire treatment approach, please refer to the informed consent provided by the MLD clinical reference centre.

A specific consent will be collected for each individual examination or invasive intervention included in the trial.

3. What risks may our child/the minor face if I/we authorise participation in the trial?

There is no risk to the newborn child in the proposed study. In fact, the residual material from the card taken for the mandatory neonatal screening will be used. The real risk to the health of the newborn comes from not participating in the study, as the patient will not be able to check whether or not he/she has MLD, a disease with an incidence of 1.1/100,000 live births in Europe (very rare).

4. How will we be informed of any unexpected results following diagnostic investigations?

Biochemical investigation for the determination of sulfatides is not expected to lead to unexpected results. However, parents/legal guardians can choose to 'not being informed' about any unexpected results that may

arise from a test, which the newborn has undergone. As for unexpected results from a genetic test possibly performed at San Raffaele Hospital, you will find detailed references to any unexpected results arising from this test in the relevant information/informed consent sheet.

5. Is it useful/necessary to inform our family doctor/paediatrician?

In view of the possible treatment that the potential screening-positive newborn may start, it is advisable to inform the family doctor/paediatrician about the study in which the infant is participating. Of course, since newborns may not have been assigned a family paediatrician yet at the time when this informed consent is administered, the paediatrician/neonatologist at the birth centre will be informed. In addition, the parents will be given a letter addressed to the general practitioner/ paediatrician: they will deliver it to the family paediatrician as soon as they are assigned one, or to a paediatrician they trust who is treating the infant.

6. What will be the commitment and responsibility of our child/the minor if I/we decide to authorise their participation? (section not longer than ½ page)

INCLUDE INFORMATION ON THE PARTICIPANT'S RESPONSIBILITIES, IN PARTICULAR:

- *Scrupulously comply with the instructions and requests of the health personnel supervising the trial and ensuring attendance at appointments.*
- *Informing the doctor supervising the trial about the following:*
  - *all the medicines the child/minor is taking, including unconventional medicines,*
  - *any side effects arising in the course of the trial,*
  - *any visits or hospitalisations in facilities other than the investigating centre,*
  - *current or previous participation in other clinical trials.*

7. Will I/we incur costs for participating in the trial? Will I/we be reimbursed for any costs? Will our child/the minor receive compensation?

No costs arising from the participation in the trial shall be borne by the patient as these are fully covered by the trial centre through the sponsor Fondazione Buzzi based on a Telethon grant.

There is also no financial compensation for participation in the trial.

8. What happens if our child/the minor suffers an injury as a consequence of participating in the trial?

The proposed study falls into the classification of non-interventional, non-drug observational studies, since participation in this study only entails performing a biochemical test in addition to those already performed for mandatory newborn screening, on a sample already taken from the newborn for this purpose. For this reason, there is no insurance cover, as provided for by the Decree of the Ministry of Labour, Health and Social Policies of 14th July 2009, on Minimum requirements for insurance policies to protect subjects participating in clinical trials of medicinal products, art. 4, paragraph 1 'The obligations under this decree do not apply to non-interventional trials (or observational studies)'.

The possible continuation of the study at the clinical reference centre for MLD, with the administration of new diagnostic tests and possible therapies, may entail inconveniences and risks that cannot be determined beforehand. For this reason, the clinical trial provides for insurance cover to protect the participation of your child/the minor, and the clinical centre itself will inform you about it.

9. How will our child's/the minor's health data, including identification data, be processed and who will have access to them during the trial?

The data, in particular personal and health data and only to the extent that it is indispensable in relation to the goal of the trial and for pharmacovigilance purposes, will be processed in accordance with EU Regulation 2016/679, known as GDPR (General Data Protection Regulation) and Legislative Decree No. 101 of 10th August 2018. In practical terms, the participant's documents will be kept in a safe place and they will not bear the patient's name in plain text. Researchers will know the name, while the documents will bear an identification code instead.

The anonymised data may be subject to control by regulatory bodies and used for scientific publications (journals, conferences).

The clinical data collected for the purpose of the trial, as well as the results of the performed examinations, will be stored for the time required by the regulations and then destroyed. It will not be destroyed only if a) it is no longer possible to trace it back to the identity of your child/the minor, because it was anonymised in the course of the trial itself; b) in the presence of your specific informed consent.
If personal data is transferred to a third country or international organisation, all protections provided for in Article 46 of GDPR 679/2016 relating to the transfer will be adopted.

Further information is included in the attached data processing authorisation form.

10. How will our child's/the minor's biological samples taken for the purpose of the trial be processed and who will have access to them?

As for health data, biological samples, which are pseudonymised (a technique that allows a natural person's personal and sensitive data to be modified and masked so that it cannot be attributed to that person directly and easily), will also be used for the purposes of the trial.

Once the trial is over, the samples will be destroyed at the end of the five-year period for samples taken for mandatory newborn screening. They will not be destroyed only if: a) it is no longer possible to trace them back to the identity of your child/the minor, because they were anonymised in the course of the trial itself, or b) in the presence of your specific informed consent and agreement with the biobank for sample storage.

11. How can I/we have access to the trial results?

Once the trial is over and data has been collected, it will be analysed to draw conclusions. The investigators undertake to make the results available to the scientific community.

The regulation provides for the possibility for the participants to access the trial results. Therefore, you may ask the investigating doctor/biologist to inform you about the general trial results.

12. Has the trial been approved by the Ethics Committee?

The LOMBARDY 1 Territorial Ethics Committee examined and approved the protocol of the proposed trial on 06th December 2023. Among other things, the Ethics Committee verified that the trial complied with the European Union's Good Clinical Practice Standards and with the ethical principles expressed in the Declaration of Helsinki, and that the safety, rights and well-being of your child/the minor were protected.

13. Whom can we contact for more information about the clinical trial in which our child/the minor has been invited to participate?

ASST FBF SACCO

Newborn Screening, Functional Genomics and Rare Diseases Unit

Dr. Cristina Cereda

Tel. 02/63635262

E-mail: cristina.cereda@asst-fbf-sacco.it

IRCCS OSPEDALE SAN RAFFAELE

Paediatric Immunohematology Unit

Dr. Francesca Fumagalli

Tel. 0226434472-4387

E-mail: fumagalli.francesca@hsr.it

14. If I/we decide to authorise participation in the trial, whom can I/we contact in case of need?

For any doubts and unplanned events during the course of the trial (doubts regarding the current treatment, side effects, decision to abandon the trial, etc.), you can contact:

ASST FBF SACCO

Newborn Screening, Functional Genomics and Rare Diseases Unit

Dr. Cristina Cereda

Tel. 02/63635262

E-mail: cristina.cereda@asst-fbf-sacco.it

IRCCS OSPEDALE SAN RAFFAELE

Paediatric Immunohematology Unit

Dr. Francesca Fumagalli

Tel. 0226434472-4387

E-mail: fumagalli.francesca@hsr.it

If you want to report to any subjects not directly involved in the trial itself any events or facts relating to the trial in which your child/the minor has participated, you may contact the Ethics Committee that approved the trial (LOMBARDY 1 Territorial Ethics Committee), the Medical Administration of the Trial Centre (ASST FBF SACCO), or the competent authority (AIFA).

15. What happens if our child/the minor comes of age during the course of the trial?

Should your child/the minor come of age during the course of the trial, he/she will become legally independent in his/her choices. Consequently, he/she will be given a new informed consent to sign.

Attachments

- 1. Overview of the study
- 2. Data processing consent form

- **3. Additional documents:** Letter for the family doctor/paediatrician

CERTIFICATION OF DELIVERY OF THE INFORMATION SHEET FOR THE PARENT(S) OR LEGAL GUARDIAN(S) TO INCLUDE A MINOR IN A CLINICAL TRIAL (PROJECT TITLE: Newborn Screening for the Early Diagnosis of Metachromatic Leukodystrophy - MLD)

_____	___/___/_____	_____	_____
Full name of doctor/biologist who delivered the information sheet	Date	Time	Signature