Development of individual therapy for cystic fibrosis using primary epithelial tissue cultures

Published: 16-11-2016
Last updated: 10-08-2024

The overall aim of the study is to improve diagnostic and therapeutic options for people with CF, especially in the context of young children and novel CFTR-targeting drugs. Primary Objective: 1. to demonstrate relations between CFTR genotype,...

Ethical review	Approved WMO
Status	Recruiting
Health condition type	Respiratory disorders congenital
Study type	Observational invasive

Summary

ID

NL-OMON43765

Source
ToetsingOnline

Brief title
Precision study

Condition

- Respiratory disorders congenital

Synonym
cystic fibrosis

Research involving
Human

Sponsors and support

Primary sponsor: Universitair Medisch Centrum Utrecht
Source(s) of monetary or material Support: NCFS

Intervention

Keyword: CFTR-targeting drugs, Cystic Fibrosis (CF), epithelial cultures, Individual therapy

Outcome measures

Primary outcome

The correlation between CFTR genotype, CFTR residual function and response to currently available therapy assessed in epithelial cells of bronchial, nasal or rectal origin in vitro, and how these parameters associate with individual in vivo clinical disease measures (FEV1, sweat chloride concentration, BMI, infection, etc).

Secondary outcome

Scientific proof of concept studies to identify potential variables for individual CF disease development and treatment efficacy:

- Characterization of CFTR DNA sequence variability (using TLA sequencing), mRNA (qRT-PCR) and protein expression (Western blotting and immunofluorescence) between patients samples.
- Characterization of epithelial barrier functions in response to pathogen challenge (CFTR function, mucus production, cilia function, cytokine production, cell differentiation and viability, pathogen killing) and upon pharmacological and genetic interventions
- Functional and genetic characterization of genetic modifiers of CF disease (e.g. alternative channel, transcription factors) using DNA sequencing (whole genome sequencing will be done for CF tissues, potential modifiers will be validated using the control population), pharmacological treatments and genetic

engineering of cells

- identification of CFTR-dependent cellular biomarkers (mRNA, protein, and metabolome profiling) using pharmacological treatments and genetic engineering of cells

Feasibility studies aiming to setup novel airway cultures and CFTR-dependent readouts.

- ability to generate and maintain airway organoid cultures and use for study of CFTR function and CF modifiers

- ability to generate and maintain submucosal gland cells in culture and use for study of CFTR function and CF modifiers.

Study description

Background summary

Cystic fibrosis is characterized by a mucosal immunodeficiency, resulting from genetic mutations in CFTR of which almost 2000 are known 1. CF disease (and response to therapy) is highly heterogeneous, which is caused by mostly unknown interactions between the
mutant CFTR, the patient-specific genetic background and the environment. For many mutations, the functional impact of the mutation on CFTR remains largely unknown. In addition, variability in CFTR expression and function in subjects with similar CFTR mutations has also been clearly observed but relations with disease severity and underlying mechanisms are mostly unknown, albeit suggested in previous studies by us and others 2-4.

CF subjects are meticulously clinically phenotyped (lung function, BMI, colonization with pathogens, bronchial alveolar lavages, CFTR function measurements in rectal biopsies of newborns or subjects with rare mutations) as to start symptomatic treatments as early as needed, all as part of standard care. Currently, new drugs that can directly restore the CFTR protein - the underlying cause of disease - cause a paradigm shift in the field 5-7. These novel treatments can achieve life-changing effects, but act in a mutation and

patient-specific fashion. The majority of subjects do not yet receive these therapies, but it is expected that many more subjects will receive these type of treatments in the near future, and that treatment should start as early as possible as to prevent disease expression. Approximately 5% of subjects in our center now receive these treatments, and a larger group (~50%) is expected to receive treatment in the near future depending on regulatory approval. As for treatment, earlier treatment is expected to most effectively limit disease.

Biomarkers that accurately predict long-term individual treatment efficacy are lacking 8,9. Current biomarkers used for these drugs are based on experiences with symptomatic treatments (e.g. change in lung function) or are individual readouts suited for CF diagnosis (e.g. sweat chloride concentration, nasal potential). Their value for CFTR targeting treatments has been demonstrated at the group level. These biomarkers can only be measured after in vivo application of drugs, and are associated with significant intra- and intersubject variation that prevent accurate measurement of individual drug efficacy 8. Especially for young children biomarkers are lacking. Novel biomarkers that facilitate a more accurate quantitation of treatment on individual CFTR function are needed, as to select and develop optimal treatment strategies in young children to prevent disease expression on the long run.

We have generated proof-of-concept that CF subjects can be preclinically selected for treatment by studying their drug response in in vitro intestinal stem cell cultures, and observed that responses of drugs in intestinal cells at the group level correlate with published outcome measures of in vivo clinical trials 2 (and Dekkers et al, 2016, Science Translational Medicine). However, we also observed that the prediction of the pulmonary response to treatment in individual patients using intestinal cells can be difficult, and we hypothesize that similar approaches using primary airway cultures may be more suited for the prediction of individual drug efficacy in the airways. A collection of paired epithelial tissue samples (rectal, nasal, bronchial) from individuals will be essential to directly compare their in vitro assay performance as well as to study their potential value for prognostic typing of individual disease severity and drug efficacy. Furthermore, in order to study intra-subject variability, we aim to collect rectal biopsies and generate intestinal organoids of 10 subjects with CF. Responses to drugs between the *new* and *old* culture of organoids will be compared.

In addition to in vitro testing of pure drugs on an individual tissue, we also observed that the activity of these drugs can be measured in plasma of patients upon treatment 10. This approach will help to study human variability in the pharmacokinetic properties of the treatment, i.e. which subjects have high circulating and low circulating levels of drugs, and how does this relate to in vivo treatment. By including plasma samples from patients, and test these before and after treatment start, we have the ability to study how the individual tissue in the lab responds to pure drugs, and how these drug responses compare to drug levels in the circulation after in vivo treatment,

and how tissues from individuals that do not receive treatment respond to circulating drug levels from subjects that already receive these drugs. This combined pharmacokinetic and pharmacodynamic modelling is already helping clinical decision-making in clinical trials and for off-label treatments of CFTR-modulating compounds, facilitating the adaptation, continuation or cessation of treatments at an individual basis. This process may significantly improve by using individual airway cultures.

We will collect airway tissue during diagnostic bronchoscopy of subjects with CF: nasal cells via brushing, bronchial cells via brushings and pulmonary biopsies. For most subjects, intestinal cultures have been already collected and biobanked in collaboration with HUB. If rectal biopsies are not available, we will collect these as well. For 10 subjects with CF with known differences in response to therapy based on previous organoid measurements, additional rectal tissue is collected to address intra-subject variability. In addition, 10 non-CF patients (0-18 yrs) will be included as a control group.
It is essential to focus on young subjects as these studies aim to develop prospective diagnostic tools for young children, and the adult CF airway epithelium is affected by long-term disease, leading to potential bias of results.

Materials are coded to study correlations between data generated in the laboratory and observations during clinical follow up in the presence or absence of treatments. People are approached to biobank surplus material for future scientific studies using a separate TcBio protocol and informed consent. Without biobank consent, all patient*s material collected for this study will be destroyed at the end of the study.

Airway epithelial stem cell cultures are generated using published protocols that allow the apparent unlimited expansion in vitro of patient-specific airway cells (from nasal and bronchial sources) 11. These culture models facilitate the study of CFTR expression and function using conventional molecular, electrophysiological and fluid secretion assays which are all operational in the lab.

We will also use the generated cell cultures and the bronchial biopsies from these subjects for proof of concept studies that aim to characterize mechanisms associated with variability in cystic fibrosis disease and response to therapy. Our previous studies in intestinal cells clearly demonstrate that the patient-specific genetic background impact the CF phenotype (e.g. swelling of organoids) and response to therapy independent of the CF-causing mutations 2 (and Dekkers et al 2016, Science Translational Medicine). In this project we focus on the functional characterization of CFTR-dependent epithelial cell functions (epithelial barrier function, electrophysiology and fluid secretion), and how these are modified by pathogens, or pharmacological and genetic interventions that modify CFTR function or (potential) modifier genes such as alternative ion channels or transcription factors that control CFTR expression.

This will involve DNA, RNA, protein and metabolic expression profiling in the context of laboratory interventions (DNA gene editing and therapy, RNA editing, and pharmacological interventions such as siRNA and small molecule treatments). It implies that we may identif

Study objective

The overall aim of the study is to improve diagnostic and therapeutic options for people with CF, especially in the context of young children and novel CFTR-targeting drugs.

Primary Objective:
1. to demonstrate relations between CFTR genotype, residual function and response to therapy in vitro using airway and intestinal biopsies and cultures, and to correlate these to clinical disease development before and after initiation of CFTR-targeting treatments.

Secondary Objective(s):
1. to establish proof of concept for novel mechanisms that contribute to disease variability between people with CF
2. to establish novel epithelial cell culture technologies for the study of CF disease

Study design

This is a monocenter, invasive observational study. 20 children with CF will be included in the study as well as 10 non-CF subjects.
The total study duration is expected to be 4 years. Study duration for each individual patient will be 1 day, consisting of 1 visit at the hospital for their bronchoscopy. An interim analysis will be performed after the first 10 subjects with CF have participated, to determine whether we are able to culture the pulmonary, nasal and intestinal materials.

Subjects who are undergoing a bronchoscopy as part of their care will be asked to participate in this study. In addition to their scopy the physician and designated study team member will collect all samples required for the conduct of this study (lung biopsies, lung brushings, nasal brushings, rectal biopsies and blood).

Subjects will be asked to store their airway tissues and cultures in the UMCU lung biobank (in progress) and the rectal organoids in HUB-CF biobank

Study burden and risks

Collecting lung and intestinal biopsies from a minor involves the risk of

temporary bleeding in a limited amount of subjects. Brushing of the nasal cavity can cause irritation of the nasal mucosa, which will disappear within a few hours. These materials provide the opportunity to develop individual-based biomarkers to investigate pulmonary disease, and individualized medicine, and is especially relevant for young children for whom these are currently lacking. Lung disease and therapy is highly heterogeneous in CF, and subject are expected to benefit most from individually tailored therapies that already start at young age. These studies provide opportunities to discover new biomarkers and drug targets for treatment of CF. Although we do not expect direct benefits of this study for the patients, the validation of airway cultures for prognostic drug efficacy testing and the potential collection of their tissue in a biobank may help to establish new diagnostic and therapeutic options for CF. At later stages, personalized treatment strategies using these biobanked materials may be realized at an improved accuracy as compared to current procedures using individual intestinal cultures.
The extension of the time under anesthesia is minimal (around 10 minutes extra). The study has a minimal risk and minimal burden in total.

Contacts

Public
Universitair Medisch Centrum Utrecht

Uppsalalaan 8
Utrecht 3584 CT
NL
Scientific
Universitair Medisch Centrum Utrecht

Uppsalalaan 8
Utrecht 3584 CT
NL

Trial sites

Listed location countries

Netherlands

Eligibility criteria

Age
Adolescents (12-15 years)
Adolescents (16-17 years)
Children (2-11 years)

Inclusion criteria

- Signed Informed Consent
- 0-18 years of age
- Two CF-causing CFTR mutations (or not known, in case of the control population)
- Undergoing a bronchoscopy for diagnostic purposes

Exclusion criteria

- CF related liver disease with abnormal coagulation

Study design

Design

Study type:	Observational invasive
Intervention model:	Other
Allocation:	Non-randomized controlled trial
Masking:	Open (masking not used)
Control:	Active
Primary purpose:	Diagnostic

Recruitment

NL

Recruitment status:	Recruiting
Start date (anticipated):	14-02-2017
Enrollment:	30
Type:	Actual

Ethics review

Approved WMO Date:	16-11-2016
Application type:	First submission
Review commission:	METC Universitair Medisch Centrum Utrecht (Utrecht)
Approved WMO Date:	11-01-2019
Application type:	Amendment
Review commission:	METC Universitair Medisch Centrum Utrecht (Utrecht)

Study registrations

Followed up by the following (possibly more current) registration

No registrations found.

Other (possibly less up-to-date) registrations in this register

No registrations found.

In other registers

Register	ID
CCMO	NL54885.041.16